

Connecting via Winsock to Dialog

Logging in to Dialog

Trying 31060000009998...Open

DIALOG INFORMATION SERVICES

PLEASE LOGON:

\*\*\*\*\*

ENTER PASSWORD:

\*\*\*\*\*

Welcome to DIALOG

Dialog level 05.27.00D

Last logoff: 13oct09 15:07:59

Logon file405 19oct09 07:47:50

\*\*\* ANNOUNCEMENTS \*\*\*

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\*\*\* FREE FILE OF THE MONTH: OCTOBER

American Business Directory (File 531)

Each month Dialog offers an opportunity to try out new or unfamiliar sources by offering \$100 of free searching (either DialUnits or connect time) in specified files. Output and Alerts charges are not included. For more details visit: <http://www.dialog.com/freelfile/> and then take a moment to get familiar with another great Dialog resource.

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EMBASE Classic (File 772) available to all customers.

NEW FILE

\*\*\*File 558, Mergent China Private Company Database

\*\*\*File 457, The Lancet(R)

FILE RENAMED

\*\*\*File 323, RAPRA: Rubber & Plastics is now RAPRA Polymer Technology

RESUMED UPDATING

\*\*\*File 523, D&B European Financial Records

\*\*\*

RELOADS COMPLETED

\*\*\*File 663, TRADEMARKSCAN(R) - Monaco

\*\*\*File 676, TRADEMARKSCAN(R) - Slovak Republic

\*\*\*File 677, TRADEMARKSCAN(R) - Liechtenstein

\*\*\*File 681, TRADEMARKSCAN(R) - Hungary

\*\*\*File 683, TRADEMARKSCAN(R) - Ireland

\*\*\*File 685, TRADEMARKSCAN(R) - Lithuania

\*\*\*File 688, TRADEMARKSCAN(R) - Portugal

\*\*\*File 697, TRADEMARKSCAN(R) - Latvia

\*\*\*

FILES REMOVED

\*\*\*File 743, New Jersey, The Record - Please use NewsRoom

\*\*\*File 301, CHEMNAME - Please use File 398 ChemSearch

\*\*\*File 388, PEDS: Defense Program Summaries

\*\*\*File 588, DMS-FI Contract Awards

\*\*\*File 559, CorpTech Directory of Technology Cos.

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>>>and events, please visit What's New from Dialog at <<<

>>><http://www.dialog.com/whatsnew/>. You can find news about <<<

>>>a specific database by entering HELP NEWS <file number>. <<<

\* \* \*

SYSTEM:HOME

Cost is in DialUnits

Menu System II: D2 version 1.8.0 term=ASCII

\*\*\* DIALOG HOMEBASE(SM) Main Menu \*\*\*

Information:

1. Announcements (new files, reloads, etc.)
2. Database, Rates, & Command Descriptions
3. Help in Choosing Databases for Your Topic
4. Customer Services (telephone assistance, training, seminars, etc.)
5. Product Descriptions

Connections:

6. DIALOG(R) Document Delivery
7. Data Star(R)

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/H = Help

/L = Logoff

/NOMENU = Command Mode

Enter an option number to view information or to connect to an online service. Enter a BEGIN command plus a file number to search a database

(e.g., B1 for ERIC).

? b 410

```

19oct09 07:47:51 User226352 Session D1178.1
$0.00 0.267 DialUnits FileHomeBase
$0.00 Estimated cost FileHomeBase
$0.00 Estimated cost this search
$0.00 Estimated total session cost 0.267 DialUnits

```

File 410:The Chronolog 2009  
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Set Items Description
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? set hi ;set hi
HIGHLIGHT set on as ''
HIGHLIGHT set on as ''
? b biochem
19oct09 07:47:57 User226352 Session D1178.2
$0.00 0.115 DialUnits File410
$0.00 Estimated cost File410
$0.02 TELNET
$0.02 Estimated cost this search
$0.02 Estimated total session cost 0.382 DialUnits

```

SYSTEM:OS - DIALOG OneSearch

```

File 5:Biosis Previews(R) 1926-2009/Oct W2
(c) 2009 The Thomson Corporation
File 6:NTIS 1964-2009/Nov W1
(c) 2009 NTIS, Intl Cpyrght All Rights Res
File 24:CSA Life Sciences Abstracts 1966-2009/Nov
(c) 2009 CSA.
File 34:SciSearch(R) Cited Ref Sci 1990-2009/Oct W2
(c) 2009 The Thomson Corp
File 40:Enviroline(R) 1975-2008/May
(c) 2008 Congressional Information Service
*File 40: This file is closed and will no longer update. For
similar data, please search File 76-Environmental Sciences.
File 41:Pollution Abstracts 1966-2009/Nov
(c) 2009 CSA.
File 45:EMCare 2009/Oct W2
(c) 2009 Elsevier B.V.
File 50:CAB Abstracts 1972-2009/Oct W2
(c) 2009 CAB International
File 65:Inside Conferences 1993-2009/Oct 16
(c) 2009 BLDSC all rts. reserv.
File 71:ELSEVIER BIOBASE 1994-2009/Oct W3
(c) 2009 Elsevier B.V.
*File 71: The file has been reloaded. Accession numbers
have changed.
File 72:EMBASE 1993-2009/Oct 19
(c) 2009 Elsevier B.V.
*File 72: The file has been synchronized to the calendar date. It
is complete and up to date as of 9/28/2009.
File 73:EMBASE 1974-2009/Oct 19

```

(c) 2009 Elsevier B.V.

\*File 73: The file has been synchronized with the calendar date.  
It is complete and up to date as of 9/28/2009.

File 76:Environmental Sciences 1966-2009/Nov  
(c) 2009 CSA.

File 98:General Sci Abs 1984-2009/Oct  
(c) 2009 The HW Wilson Co.

File 103:Energy SciTec 1974-2009/Oct B1  
(c) 2009 Contains copyrighted material

\*File 103: For access restrictions see Help Restrict.

File 136:BioEngineering Abstracts 1966-2007/Jan  
(c) 2007 CSA.

\*File 136: This file is closed.

File 143:Biol. & Agric. Index 1983-2009/Sep  
(c) 2009 The HW Wilson Co

File 144:Pascal 1973-2009/Oct W3  
(c) 2009 INIST/CNRS

File 154:MEDLINE(R) 1990-2009/Oct 16  
(c) format only 2009 Dialog

File 155:MEDLINE(R) 1950-2009/Oct 16  
(c) format only 2009 Dialog

File 156:ToxFile 1965-2009/Oct W2  
(c) format only 2009 Dialog

File 162:Global Health 1983-2009/Oct W2  
(c) 2009 CAB International

File 172:EMBASE Alert 2009/Oct 19  
(c) 2009 Elsevier B.V.

\*File 172: The file has been synchronized with today's calendar date.  
It  
is complete and up to date.

File 305:Analytical Abstracts 1980-2009/Aug W4  
(c) 2009 Royal Soc Chemistry

\*File 305: Alert feature enhanced for multiple files, duplicate  
removal, customized scheduling. See HELP ALERT.

File 369:New Scientist 1994-2009/Oct W2  
(c) 2009 Reed Business Information Ltd.

File 370:Science 1996-1999/Jul W3  
(c) 1999 AAAS

\*File 370: This file is closed (no updates). Use File 47 for more  
current  
information.

File 393:Beilstein Database - Abstracts 2008/Q2  
(c) 2008 Beilstein GmbH

File 399:CA SEARCH(R) 1967-2009/UD=15116  
(c) 2009 American Chemical Society

\*File 399: Use is subject to the terms of your user/customer  
agreement.

IPCR/8 classification codes now searchable as IC=. See HELP NEWSIPCR.

File 434:SciSearch(R) Cited Ref Sci 1974-1989/Dec  
(c) 2006 The Thomson Corp

Set Items Description

```

    ---  -----
? s amazonensis and promastigote and surface and antigen?
Processing
Processed 20 of 29 files ...
Completed processing all files
      12167 AMAZONENSIS
      14701 PROMASTIGOTE
      8592688 SURFACE
      4820606 ANTIGEN?
S1      164 AMAZONENSIS AND PROMASTIGOTE AND SURFACE AND
ANTIGEN?
? rd s1

```

>>>Duplicate detection is not supported for File 393.

```

>>>Records from unsupported files will be retained in the RD set.
S2      51 RD S1 (unique items)
? s amazonensis and ((promastigote and surface and antigen?) or PSA
or ESA)
Processing
Processed 20 of 29 files ...
Completed processing all files
      12167 AMAZONENSIS
      14701 PROMASTIGOTE
      8592688 SURFACE
      4820606 ANTIGEN?
      136324 PSA
      40318 ESA
S3      175 AMAZONENSIS AND ((PROMASTIGOTE AND SURFACE AND
ANTIGEN?)
OR PSA OR ESA)
? rd s3

```

>>>Duplicate detection is not supported for File 393.

```

>>>Records from unsupported files will be retained in the RD set.
S4      55 RD S3 (unique items)
? ds

Set      Items      Description
S1      164      AMAZONENSIS AND PROMASTIGOTE AND SURFACE AND ANTIGEN?
S2      51      RD S1 (unique items)
S3      175      AMAZONENSIS AND ((PROMASTIGOTE AND SURFACE AND
ANTIGEN?) OR
PSA OR ESA)
S4      55      RD S3 (unique items)
? s ((promastigote and surface and antigen?) or PSA or ESA)
Processing
Processed 20 of 29 files ...
Completed processing all files
      14701 PROMASTIGOTE
      8592688 SURFACE

```

```

4820606 ANTIGEN?
136324 PSA
40318 ESA
S5 177478 ((PROMASTIGOTE AND SURFACE AND ANTIGEN?) OR PSA OR
ESA)
? s s5 and (leishmania or amazonensis)
177478 S5
159840 LEISHMANIA
12167 AMAZONENSIS
S6 1112 S5 AND (LEISHMANIA OR AMAZONENSIS)
? rd s6

```

>>>Duplicate detection is not supported for File 393.

>>>Records from unsupported files will be retained in the RD set.

```

S7 335 RD S6 (unique items)
? s s7 nor PY>2004

```

>>>Term "NOR" in invalid position

```

? s s7 not PY>2004

```

Processing

Processed 10 of 29 files ...

Completed processing all files

```

335 S7
37837515 PY>2004
S8 298 S7 NOT PY>2004
? ds

```

Set	Items	Description
S1	164	AMAZONENSIS AND PROMASTIGOTE AND SURFACE AND ANTIGEN?
S2	51	RD S1 (unique items)
S3	175	AMAZONENSIS AND ((PROMASTIGOTE AND SURFACE AND ANTIGEN?) OR

PSA OR ESA)

```

S4 55 RD S3 (unique items)
S5 177478 ((PROMASTIGOTE AND SURFACE AND ANTIGEN?) OR PSA OR
ESA)
S6 1112 S5 AND (LEISHMANIA OR AMAZONENSIS)
S7 335 RD S6 (unique items)
S8 298 S7 NOT PY>2004

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```

? s s8 and 42.5

```

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298 S8
0 42.5

```

```

S9 0 S8 AND 42.5

```

```

? s s8 and kda

```

```

298 S8
958548 KDA

```

```

S10 52 S8 AND KDA

```

```

? t s10/7/all

```

>>>Format 7 is not valid in file 143

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10/7/1 (Item 1 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

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17674704 BIOSIS NO.: 200400045461

Immunolocalization of Leishmania (Viannia) braziliensis membrane antigens recognized by mAbs SST-2, SST-3, and SST-4.

AUTHOR: Silveira T G V; Takahashi H K; Straus A H (Reprint)

AUTHOR ADDRESS: Department of Biochemistry, Universidade Federal de Sao

Paulo/Escola Paulista de Medicina, Rua Botucatu 862, Sao Paulo, SP, 04023-900, Brazil\*\*Brazil

AUTHOR E-MAIL ADDRESS: straus.bioq@epm.br

JOURNAL: Parasitology 127 (5): p449-456 November 2003 2003

MEDIUM: print

ISSN: 0031-1820

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: The immunolocalization of Leishmania (Viannia) braziliensis stage-specific antigens recognized by mAbs was analysed by transmission electron microscopy. The antigen recognized by mAb SST-2 was present at the surface of promastigotes, including the flagellum and flagellar pocket. The reactivity of SST-2 with isolates of

different serodemes showed a pronounced microheterogeneity in terms of

the number of reactive bands within the low molecular weight range from

24 to 33 kDa. The 180 kDa glycoprotein recognized by mAb

SST-3 was present only in the flagellar membrane. SST-3 also recognized

multiple discrete bands from 160 to 200 kDa, as observed in several serodemes. In contrast, mAb SST-4, which recognizes a 98 kDa antigen, showed weak labelling on the promastigote surface by transmission electron microscopy and indirect immunofluorescence. Based on Western blotting, indirect immunofluorescence, and solid-phase radioimmunoassay, the antigens recognized by mAbs SST-2, SST-3 and SST-4 were present in all L.

(V.)

braziliensis analysed, from 7 different serodemes.

10/7/2 (Item 2 from file: 5)

DIALOG(R)File 5:Biosis Previews(R)

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17657659 BIOSIS NO.: 200400028416

Immune complex antigens as a tool in serodiagnosis of kala-azar.

AUTHOR: Chakraborti Tapati (Reprint); Sarkar Dwijen; Ghosh Dilip K

AUTHOR ADDRESS: Department of Biochemistry and Biophysics, University of

Kalyani, Kalyani, WB, 741235, India\*\*India

AUTHOR E-MAIL ADDRESS: tchakraborti@yahoo.com  
JOURNAL: Molecular and Cellular Biochemistry 253 (1-2): p191-198  
November  
2003 2003  
MEDIUM: print  
ISSN: 0300-8177 \_(ISSN print)  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: English

ABSTRACT: The 63 kDa surface antigen of Leishmania promastigotes is one of the most important virulent factors in establishing the host parasite relationship. This glycoprotein is revealed by surface iodination study as well as by metabolic labeling and immunoblot methods. In search of this specific antigen for serodiagnosis, immune complexes (ICs) were isolated from kala-azar

patient sera and analysed by SDS-PAGE and Western immunoblotting. The

immunoblot of kala-azar IC with patient sera, anti-promastigote sera and anti gp63 sera detected the major antigen of 55 kDa. This recognition suggests that 55 kDa antigen and gp63 have common antigenic epitope(s). Normal IC did not react with anti gp63 sera indicating absence of this antigen in normal IC. To confirm the parasitic origin of the 55 kDa antigen of kala-azar IC, in vitro IC was formed with parasite antigen and acid dissociated kala-azar IC antibody. This indicated the antigenic similarity of the 55 kDa antigen and gp63 antigen of the parasite. This also suggested that the former antigen may have been processed from gp63. In summary, identification of parasite antigen (55 kDa) in IC of kala-azar patients' sera may be useful in developing a serodiagnostic assay of visceral leishmaniasis. Several other antigens are visualized in kala-azar IC when developed with patient sera. But specificity and efficacy of these antigens have not yet been evaluated in serodiagnosis of the disease.

10/7/3 (Item 3 from file: 5)  
DIALOG(R)File 5:Biosis Previews(R)  
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17626210 BIOSIS NO.: 200300576887  
gp63 homologues in Trypanosoma cruzi: Surface antigens with metalloprotease activity and a possible role in host cell infection.  
AUTHOR: Cuevas Ileana C; Cazzulo Juan J; Sanchez Daniel O (Reprint)  
AUTHOR ADDRESS: Instituto de Investigaciones Biotecnologicas, Universidad

Nacional de General San Martin, INTI, Edificio 24, 1650, San Martin, Buenos Aires, Argentina\*\*Argentina  
AUTHOR E-MAIL ADDRESS: dsanchez@iib.unsam.edu.ar  
JOURNAL: Infection and Immunity 71 (10): p5739-5749 October 2003 2003  
MEDIUM: print

ISSN: 0019-9567 \_(ISSN print)  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: English

ABSTRACT: gp63 is a highly abundant glycosylphosphatidylinositol (GPI)-anchored membrane protein expressed predominantly in the promastigote but also in the amastigote stage of *Leishmania* species. In *Leishmania* spp., gp63 has been implicated in a number of steps in establishment of infection. Here we demonstrate that *Trypanosoma cruzi*, the etiological agent of Chagas' disease, has a family of gp63 genes composed of multiple groups. Two of these groups, Tcgp63-I and -II, are present as high-copy-number genes. The genomic organization and mRNA expression pattern were specific for each group. Tcgp63-I was widely expressed, while the Tcgp63-II group was scarcely detected in Northern blots, even though it is well represented in the *T. cruzi* genome. Western blots using sera directed against a synthetic peptide indicated that the Tcgp63-I group produced proteins of approx 78 kDa, differentially expressed during the life cycle. Immunofluorescence staining and phosphatidylinositol-specific phospholipase C digestion confirmed that Tcgp63-I group members are surface proteins bound to the membrane by a GPI anchor. We also demonstrate the presence of metalloprotease activity which is attributable, at least in part, to Tcgp63-I group. Since antibodies against Tcgp63-I partially blocked infection of Vero cells by trypomastigotes, a possible role for this group in infection is suggested.

10/7/4 (Item 4 from file: 5)  
DIALOG(R)File 5:Biosis Previews(R)  
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17611265 BIOSIS NO.: 200300579984  
Surface glycoprotein PSA (GP46) expression during short- and long-term culture of *Leishmania chagasi*.  
AUTHOR: Beetham Jeffrey K (Reprint); Donelson John E; Dahlin Rebecca R  
AUTHOR ADDRESS: Department of Veterinary Pathology, Iowa State University,  
2714 Veterinary Medicine, Ames, IA, 50011, USA\*\*USA  
AUTHOR E-MAIL ADDRESS: jbeetham@iastate.edu  
JOURNAL: Molecular & Biochemical Parasitology 131 (2): p109-117  
October  
2003 2003  
MEDIUM: print  
ISSN: 0166-6851  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: The mRNAs encoding promastigote surface antigen (PSA) of *Leishmania chagasi* have previously been shown to increase about 30-fold as in vitro cultured parasites progress from logarithmic to stationary phase, growth phases that are, respectively associated with parasites having low and high infectivity to mammals. Experiments reported here establish by western blot analysis that PSA proteins of 44 and 66 kDa also increase about 30-fold as parasite cultures reach stationary phase. Serial passage of parasite cultures resulted in a progressive reduction in PSA protein and RNA abundance to levels less than 3% that of cultures newly-initiated with parasites derived from a parasitized rodent. Loss of PSA mRNA abundance in serially passaged cells was not due to reduced PSA gene transcription rates, as determined by nuclear run-on assays. Neither was the loss associated with a marked decrease in PSA mRNA stability. Analysis of PSA RNA stability in the presence of actinomycin D, an inhibitor of transcription elongation, failed to detect a difference in fully processed cytosolic PSA mRNA stability regardless of the number of times a culture was passaged or the growth phase of the culture. Based on the lack of detectable difference in (cytosolic) mature PSA mRNA stability during promastigote development, the data indirectly suggest that the regulated expression of PSA in cells from low-passage cultures and the loss of PSA expression in high-passage cultures may be mediated by nuclear events that occur after transcription of the PSA genes and before arrival of the mature mRNAs in the cytoplasm.

10/7/5 (Item 5 from file: 5)  
DIALOG(R)File 5:Biosis Previews(R)  
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16745135 BIOSIS NO.: 200200338646  
Comparison of the post-transcriptional regulation of the mRNAs for the surface proteins PSA (GP46) and MSP (GP63) of *Leishmania chagasi*

AUTHOR: Myung Karen S; Beetham Jeffrey K; Wilson Mary E; Donelson John E

(Reprint)

AUTHOR ADDRESS: Department of Biochemistry, University of Iowa, Iowa City, IA, 52242, USA\*\*USA

JOURNAL: Journal of Biological Chemistry 277 (19): p16489-16497 May 10,

2002 2002  
MEDIUM: print  
ISSN: 0021-9258  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: English

ABSTRACT: MSP (GP63) and PSA (GP46) are abundant 63- and 46-kDa glycolipid-anchored proteins on the surface of the promastigote form of most Leishmania species. MSP is a zinc metalloprotease that confers resistance to host complement-mediated lysis. PSA contains internal repeats of 24 amino acids, and its function is unknown. The steady state levels of mRNAs for both glycoproteins are regulated post-transcriptionally, resulting in about a 30-fold increase as Leishmania chagasi promastigotes grow in vitro from logarithmic phase to stationary phase. Previous studies showed the 3'-untranslated regions (3'-UTRs) of these mRNAs are essential for this post-transcriptional regulation. These two 3'-UTRs of 1.0 and 1.3 kilobases were cloned immediately down-stream of a beta-galactosidase reporter gene in a plasmid, and segments were systematically deleted to examine which portions of the 3'-UTRs contribute to the post-transcriptional regulation. The 92-nucleotide segment of greatest similarity between the two 3'-UTRs was deleted without loss of regulation, but the segments flanking this similarity region have positive regulatory elements essential for the regulation. We propose that similar, but non-identical, molecular mechanisms regulate the parallel expression of these two L. chagasi mRNAs despite their lack of sequence identity. These post-transcriptional mechanisms resemble the mechanism recently suggested for the regulation of mRNAs encoding the dipeptide (EP) and pentapeptide (GPEET) repeat proteins in Trypanosoma brucei that involves interactions between positive and negative regulatory elements in the 3'-UTR.

10/7/6 (Item 6 from file: 5)  
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16383340 BIOSIS NO.: 200100555179  
Inhibition of macrophage invasion by monoclonal antibodies specific to Leishmania (Viannia) braziliensis promastigotes and

characterisation of their antigens  
AUTHOR: Silveira Thais G V; Suzuki Erika; Takahashi Helio K; Straus  
Anita H  
(Reprint)  
AUTHOR ADDRESS: Department of Biochemistry, Universidade Federal de  
Sao  
Paulo/Escola Paulista de Medicina, Rua Botucatu 862, Sao Paulo, SP,  
04023-900, Brazil\*\*Brazil  
JOURNAL: International Journal for Parasitology 31 (13): p1451-1458  
November, 2001 2001  
MEDIUM: print  
ISSN: 0020-7519  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: English

ABSTRACT: Monoclonal antibodies that specifically recognise  
Leishmania (Viannia) braziliensis promastigotes were produced and  
termed SST-2, SST-3 and SST-4. SST-2 recognises a conformational  
epitope  
present in a 24-28 kDa doublet and in a 72 kDa component, as  
verified by Western blotting. Indirect immunofluorescence showed  
that the  
antigen recognised by SST-2 is distributed homogeneously on the  
parasite surface. SST-3 recognises a flagellar glycoprotein of  
approx 180 kDa. The reactivity of this mAb was abolished by sodium  
m-periodate treatment, indicating that SST-3 reacts with a  
carbohydrate  
epitope of the 180 kDa antigen. SST-4 recognises a  
conformational epitope of a 98 kDa antigen. SST-2, SST-3 and  
SST-4 were specific to L. (V.) braziliensis promastigote forms.  
Indirect immunofluorescence did not show reactivity of SST-2 or  
SST-3  
with amastigotes of L. (V.) braziliensis, or with promastigotes of  
Leishmania (Viannia) panamensis, Leishmania (Viannia)  
guyanensis, Leishmania (Viannia) naiffi, Leishmania (Viannia)  
lainsoni, Leishmania (Leishmania) amazonensis,  
Leishmania (Leishmania) major, or Leishmania (  
Leishmania) chagasi. We also evaluated the involvement of SST-2,  
SST-3 and SST-4 antigens in parasite-macrophage interaction. Fab  
fragments of SST-3 and SST-4 significantly inhibited the  
infectivity of  
L. (V.) braziliensis promastigotes to mouse peritoneal macrophages.

10/7/7 (Item 7 from file: 5)  
DIALOG(R)File 5:Biosis Previews(R)  
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14658075 BIOSIS NO.: 199800452322  
Leishmania major: Cell type dependent distribution of a 43 kDa  
antigen related to silent information regulatory-2 protein family

AUTHOR: Zemzoumi Khalid; Sereno Denis; Francois Celine; Guilvard Elaine;

Lemesre Jean-Loup; Ouaisi Ali (Reprint)

AUTHOR ADDRESS: CJF INSERM 96-04, Centre ORSTOM Montpellier, 911 Ave.

Agropolis, BP 5045, 34032 Montpellier Cedex 01, France\*\*France

JOURNAL: Biology of the Cell (Paris) 90 (3): p239-245 June, 1998 1998

MEDIUM: print

ISSN: 0248-4900

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: In previous studies we have characterized several Leishmania major polypeptides and showed that one member of this group (LmSIR2rp) shared significant homology to silent information regulator 2 (SIR2) of *Saccharomyces cerevisiae*, a protein playing a role

in both telomeric and mating type loci repression in these organisms. In

the present study, by using molecular and immunological approaches, we

could identify LmSIR2rp homologues in different Leishmania species and developmental stages (eg logarithmic (LP) and stationary phase promastigotes (SP) and amastigotes). The reactive antigen was also detected in *Trypanosoma cruzi* extracts. Surprisingly, immunofluorescence

assays revealed that LmSIR2rp is associated mainly with cytoplasmic granules of different sizes and numbers depending on the life stage of

the parasite used. No reactivity was observed in the nucleus, in agreement with the Western blot showing an absence of

immunoreactivity of

anti-LmSIR2rp immune serum against parasite nuclear extracts.

Furthermore, immunoprecipitation of (35S)methionine-labeled promastigote antigens after pulse chase experiments, using anti-LmSIR2rp fusion protein antibodies, showed that the protein is among

parasite excreted-secreted antigens (ESA). Moreover, immunofluorescence assays conducted with short time incubations of either

purified LmSIR2rp or viable promastigotes with murine macrophages, revealed that LmSIR2rp could be bound to the macrophage surface.

The unexpected cytoplasmic localization of LmSIR2rp and its presence in

ESA may suggest a new mode of action for silent information regulatory factor homologues.

10/7/8 (Item 8 from file: 5)

DIALOG(R)File 5:Biosis Previews(R)

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13031893 BIOSIS NO.: 199598499726

Expression of antigens in virulent and avirulent Indian strains of  
Leishmania donovani

AUTHOR: Sanyal Tapati; Gangopadhyay Prosenjit; Ghosh Dilip K; Sarkar  
Dwijen

(Reprint)

AUTHOR ADDRESS: Dep. Cell Biology, Indian Inst. Chemical Biology, 4  
Raja S.

C. Mullick Road, Calcutta 700 032, India\*\*India

JOURNAL: Journal of Biosciences (Bangalore) 19 (3): p291-299 1994  
1994

ISSN: 0250-5991

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: Indo-Gen mediated surface labelling with 125I demonstrated  
differences in surface oriented antigens between virulent and  
avirulent promastigotes of Leishmania donovani. In case of virulent  
strains, surface polypeptides with molecular masses of 63, 53, 42  
and 38 kDa were found to be labelled with 125I whereas in the case  
of avirulent strains 68, 55, 50, 46, 42 and 33 kDa components were  
iodinated. Further studies by immunoblot assay using different  
subcellular fractions of virulent and avirulent parasites

demonstrated

that antibody raised against gp63 cross-reacted with the 63 and 60  
kDa antigen of the virulent and avirulent Leishmania

donovani strains of Indian origin respectively. It indicates that  
these

two polypeptides are antigenically similar. When virulent and  
avirulent cells were grown in the presence of varying concentration  
of

tunicamycin and immunoblotted with anti gp63, it was observed that  
with

increasing concentration of tunicamycin the 63 kDa polypeptide of  
the virulent cells shifted to approximately 58-57 kDa and the 60  
kDa polypeptide of the avirulent cells shifted to 57 kDa.

This suggests that glycosylation may play an important role in  
antigenic variation between virulent and avirulent parasites.

10/7/9 (Item 9 from file: 5)

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12817543 BIOSIS NO.: 199598285376

Detection of serum antibodies against Leishmania 94 kDa

antigen in visceral and cutaneous leishmaniasis due to  
Leishmania infantum

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JOURNAL: Parasite 2 (1): p13-21 1995 1995  
ISSN: 1252-607X  
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RECORD TYPE: Abstract  
LANGUAGE: English

ABSTRACT: Leishmania promastigotes polypeptides are analyzed by immunoblotting with sera from patients infected with different Leishmania species and presenting visceral or cutaneous infections. These sera recognize Leishmania polypeptides in several molecular masses. The major findings of this study are as follow. 1) The Leishmania 94 kDa antigen, which is specifically recognized by all sera from L. infantum-infected patients with visceral infection, is recognized by some sera from L. infantum-infected patients presenting cutaneous infection. 2) All patients with cutaneous infections due to L. tropica, L. amazonensis, or L. guyanensis do not develop anti-94 kDa antibodies, whatever the Leishmania species used as antigens. 3) Difference in electrophoretic mobilities is seen between the 94 kDa antigen identified by sera from Leishmania infantum-infected patients, and the antigen both recognized by the Concanavalin A lectin and a rabbit antiserum raised against deglycosylated Promastigote Surface Protease.

10/7/10 (Item 10 from file: 5)  
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12542091 BIOSIS NO.: 199598009924  
The Kinetoplastid Membrane Protein 11 of Leishmania donovani and African Trypanosomes Is a Potent Stimulator of T-Lymphocyte Proliferation  
AUTHOR: Tolson Douglas L; Jardim Armando; Schnur Lionel F; Stebeck Caroline  
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JOURNAL: Infection and Immunity 62 (11): p4893-4899 1994 1994  
ISSN: 0019-9567  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: English

ABSTRACT: Kinetoplastid membrane protein 11 (KMP-11) from Leishmania donovani is an abundant 11-kDa surface membrane glycoprotein. Lymph node cells from mice of six different H-2 haplotypes immunized with

KMP-11 or with *L. donovani* promastigotes were stimulated to proliferate in vitro with purified KMP-11. Primed purified T cells required antigen presentation since they were not stimulated unless KMP-11-pulsed or *L. donovani*-infected macrophages were added. Promastigotes of a wide variety of *Leishmania* species and procyclic forms of African trypanosomes stimulated proliferation of KMP-11-primed or *L. donovani* promastigote-primed lymph node cells. All of the *Leishmania* promastigotes and African trypanosomes tested contained an 11-kDa protein, as detected by immunoblotting with KMP-11-specific monoclonal antibodies. The widespread distribution of the 11-kDa (KMP-11) molecules and their ability to stimulate strong T-lymphocyte proliferation in a non-H-restricted fashion suggest that they may be important molecules for induction of cell-mediated immune responses.

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12069197 BIOSIS NO.: 199497090482  
Relationships between cell surface protease and acid phosphatase activities of *Leishmania* promastigote  
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JOURNAL: Molecular Biology Reports 18 (3): p189-195 1993 1993  
ISSN: 0301-4851  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: English

ABSTRACT: A correlation between the ratio of the cell surface protease activity to phosphatase activity and the complexity of the pattern of cell surface exposed polypeptides of *Leishmania* promastigotes was demonstrated for various strains grown under similar conditions. The ratio of the cell surface protease activity to acid phosphatase activity was high for *L. major* and *L.b. panamensis* and it correlates with the expression of a single polypeptide of 63 KDa on their cell surface. Intermediate and lower ratios of these enzymatic activities relate with more complex radioiodinated patterns: two

main bands in *L.b. guyanensis* (70 and 58 KDa) and *L.b. braziliensis* (72 and 60 KDa) and three main bands 65, 50, 27 KDa in all *L.m. mexicana* strains tested. Evidence is presented that the acid phosphatase located on the *L.m. mexicana* cell surface is not an artifact due to a secondary absorption of the secreted acid phosphatase from the culture medium. These results confirm the *Leishmania* antigen cell surface heterogeneity. The implications on the biology of *Leishmania* and the clinical manifestation of leishmaniasis are discussed.

10/7/12 (Item 12 from file: 5)  
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11972681 BIOSIS NO.: 199396137097  
Antigen-reactive gamma-delta T cells in human leishmaniasis  
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JOURNAL: Journal of Immunology 151 (7): p3712-3718 1993  
ISSN: 0022-1767  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: English

ABSTRACT: The importance of Ag-specific gamma-delta T lymphocytes in human immune responses to pathogenic organisms is unknown. In the present study the expression of gamma-delta TCR on T lymphocytes from patients with cutaneous, mucosal, or visceral leishmaniasis was examined. All of these patient groups had elevated levels of gamma-delta T cells in peripheral blood. Patients' gamma-delta T cells included CD8+ as well as null cells. The percentage of T cells expressing gamma-delta TCR was increased significantly by stimulation in vitro with certain parasite Ag. T-cell lines generated by stimulation with promastigote lysates of *Leishmania amazonensis* or *L. braziliensis* typically contained 25 to 60% gamma-delta T cells. In contrast, two immunodominant surface Ag of *L. amazonensis*, gp63 and gp42, did not expand gamma-delta T cells from infected patients, although both Ag elicited strong alpha-beta T-cell responses. gamma-delta T cells isolated from a

Leishmania-specific T-cell line responded to stimulation with promastigote lysate. Of particular interest, gamma-delta T cells from PBMC of a patient with mucosal leishmaniasis responded to stimulation with a recombinant 70 kDa heat shock protein of *L. chagasi*. This study demonstrated that several clinical forms of leishmaniasis induced elevated numbers of gamma-delta T cells that responded specifically to *Leishmania* Ag in vitro. Therefore, this component of the T-cell response to *Leishmania* may impact the outcome of clinical disease.

10/7/13 (Item 13 from file: 5)  
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11841168 BIOSIS NO.: 199396005584  
Leishmania donovani surface glycoconjugate GP36 is the major immunogen component of the fucose-mannose ligand (FML)  
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JOURNAL: Acta Tropica 53 (1): p59-72 1993  
ISSN: 0001-706X  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: English

ABSTRACT: *Leishmania donovani* promastigote glycoconjugate ligands, studied in our laboratory, that interact with the internalization receptors on BALB/c macrophages: the 'fucose mannose ligand' (FML), the 'phosphate mannogalactan ligand' (PMGL), and the 'lipopeptidophosphoglycan' (LPPD), interfered also with interaction between amastigotes and host cells in vitro. Among the three compounds studied, the FML was shown to be the most potent inhibitor of both promastigote and amastigote internalization, and to be present on parasite surface during the vertebrate-host cycle. The FML, but not the other two glycoconjugates, is a potent immunogen in rabbits (ELISA, agglutination and immuno-blots). Rabbit hyperimmune sera recognized essentially the 36 kDa band of FML. Mouse monoclonal antibodies against FML recognized either the 36 kDa or the 55 kDa band. No cross-reactivity between these two FML components was detected. No antigenic similarity could be detected between the 36 and 59 kDa bands of FML and the 'GP63' (promastigote surface proteinase) major surface leishmanial antigen. The 36 kDa-glycoprotein was identified as the major FML antigenic fraction and designated 'GP36'. The integrity of the glycidic moiety was

necessary for its antigenicity. This *L. donovani* surface glycoprotein is apparently one of the major molecules involved in interactions between the parasite and the vertebrate host.

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11409769 BIOSIS NO.: 199294111610  
IDENTIFICATION OF A GP63 SURFACE GLYCOPROTEIN IN LEISHMANIA  
-TARENTOLAE  
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JOURNAL: FEMS Microbiology Letters 96 (1): p89-92 1992  
ISSN: 0378-1097  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: ENGLISH

ABSTRACT: The promastigote stage of most if not all *Leishmania* species possesses an abundant surface glycoprotein of 63 kDa (gp63) that has protease activity. We show that the lizard parasite *Leishmania tarentolae* appears to lack the surface protease activity. *L. tarentolae* does, however, possess an approximately 63-kDa molecule that is antigenically cross-reactive with the *L. major* gp63. Additionally, the genome of *L. tarentolae* contains sequences that  
hybridise at high stringency to a *L. major* gp63 gene probe.

10/7/15 (Item 15 from file: 5)  
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11174942 BIOSIS NO.: 199293017833  
STRUCTURE AND ANTIGENICITY OF THE LIPOPHOSPHOGLYCAN FROM  
LEISHMANIA-MAJOR AMASTIGOTES  
AUTHOR: MOODY S F (Reprint); HANDMAN E; BACIC A  
AUTHOR ADDRESS: WALTER AND ELIZA HALL INST MED RES, IMMUNOPARASITOL  
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JOURNAL: Glycobiology 1 (4): p419-424 1991  
ISSN: 0959-6658  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: ENGLISH

ABSTRACT: The lipophosphoglycan (LPG) of the intracellular amastigote form

of the protozoan parasite *Leishmania major* is chemically distinct from the LPG on the surface of the extracellular promastigote form. Amastigote LPG is composed of the monosaccharides galactose, glucose, mannose, glucosamine and inositol in the molar ratio 51:30:24:1:1; arabinose is absent. The lipid anchor comprises four alkylglycerols, with alkyl chain lengths 24:0, 22:0, 20:0 and 26:0 in the molar ratio 68:18:8:6. Phosphate is present at 4% w/w of total carbohydrate. HPLC gel permeation reveals LPG to be a polydisperse family of molecules Mr 100-6 kDa. The results from immunological studies with LPG-directed antibodies are consistent with amastigote LPG having the expected tripartite structure of GPI-anchor, a core glycan and the phosphorylated disaccharide repeat backbone. Human sera from *L. major* patients bound amastigote LPG in enzyme-linked immunosorbent assays.

10/7/16 (Item 16 from file: 5)  
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10315683 BIOSIS NO.: 199090100162  
TRYPANOSOMA-CRUZI DIFFERENTIAL EXPRESSION AND DISTRIBUTION OF AN 85-KDA POLYPEPTIDE EPITOPE BY IN-VITRO DEVELOPMENTAL STAGES  
AUTHOR: OUAISSI M A (Reprint); DUBREMETZ F J; KUSNIERZ J P; CORNETTE J;  
LOYENS M; TAIBI A; MARTY B; VELGE P; RIZVI F; CAPRON A  
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JOURNAL: Experimental Parasitology 71 (2): p207-217 1990  
ISSN: 0014-4894  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: ENGLISH

ABSTRACT: The expression by *Trypanosoma cruzi* developmental stages of an 85-kDa polypeptide epitope defined by the 155D3 monoclonal antibody (mAb) has been investigated. Immunoprecipitation revealed the presence of an 85-kDa antigen in the NP-40 soluble extract of parasites freshly released from infected fibroblasts; this antigen was not found in epimastigote and *Leishmania infantum* promastigote. Indirect immunofluorescence revealed that the mAb 155D3 failed to react with trypomastigotes, whereas extracellular amastigotes were heavily stained. Positive organisms displayed either surface or polar

fluorescence. Since the same mAb immunoprecipitated the 85-kDa antigen in both radioactive iodine- and methionine-labeled trypomastigote detergent soluble extracts, the reactive epitope is likely to be hidden in a cryptic site in trypomastigotes. An alternative explanation for the negative immunofluorescence on trypomastigotes and the positive immunoprecipitation is the presence, in the extracts, of a small population of parasites already expressing the 155D3 epitope. Immunoelectron microscopy revealed that the target epitope is heterogenously distributed among the populations of differentiating parasites. Two types of immunogold labeling were observed: (a) mAb revealed a high amount of reactive material associated with the periphery of the parasites and (b) a label was observed on the inner surface of peripheral vacuoles that might correspond to cross sections of inflated flagellar pockets and in association with vesicles which were released by the parasites. The surface expression of the epitope recognized by the 155D3 mAb was followed by fluorescence-activated cell-sorting analysis. The results showed that the epitope is increasingly accessible during trypomastigote differentiation in vitro. Taken together, these results suggest that the epitope reacting with the 155D3 mAb is heavily expressed on extracellular amastigotes after the transformation process and, thus, appears to be developmentally regulated.

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10187195 BIOSIS NO.: 199089105086  
IDENTIFICATION OF CYTOPLASMIC SOLUBLE ANTIGENS RELATED TO THE MAJOR SURFACE ANTIGENS OF LEISHMANIA -BRAZILIENSIS-BRAZILIENSIS AND LEISHMANIA-DONOVANI-CHAGASI  
AUTHOR: KUTNER S (Reprint); PELLERIN P; BRENIERE S F  
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JOURNAL: Parasitology Research 76 (3): p185-191 1990  
ISSN: 0932-0113  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: ENGLISH

ABSTRACT: This study describes the identification of aqueous-soluble antigens in Leishmania promastigotes immunologically and

biochemically closely related to the major surface antigen. Proteins from surface-iodinated *L. braziliensis braziliensis* and *L. donovani chagasi* promastigotes, extracted and separated by partitioning in the detergent Triton X-114, were analyzed. Immunoblotting of the extracted proteins, using homologous antisera, showed recognition of a 72-kDa labeled, amphiphilic antigen of *L. b. braziliensis* and a 65-kDa surface antigen of *L. d. chagasi*. The respective homologous sera also recognized non-labeled hydrophilic antigens, similar in their apparent molecular weights to the major surface antigens. The amphiphilic and hydrophilic antigens of each species were found to share common antigens determinants, inasmuch as monospecific antibodies that recognized the amphiphilic protein reacted with the hydrophilic antigen. Structural homology was also obtained in the peptide-digestion profiles of the amphiphilic and the respective hydrophilic major antigens. Zymogram assay showed that both amphiphilic and hydrophilic fractions displayed proteolytic activity that could be directly attributed to the major *L. b. braziliensis* and *L. d. chagasi* antigens. The hydrophilic antigens found in this study are probably not hydrolytic products of the surface antigens and occur in large quantities in the promastigote cytosol.

10/7/18 (Item 18 from file: 5)  
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10187143 BIOSIS NO.: 199089105034  
THE MAJOR SURFACE GLYCOPROTEIN GP63 IS PRESENT IN BOTH LIFE STAGES OF LEISHMANIA  
AUTHOR: FROMMEL T O (Reprint); BUTTON L L; FUJIKURA Y; MCMASTER W R  
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JOURNAL: Molecular and Biochemical Parasitology 38 (1): p25-32 1990  
ISSN: 0166-6851  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: ENGLISH

ABSTRACT: Leishmania exist as extracellular promastigotes which multiply in the gut of the sandfly insect vector and as intracellular amastigotes which divide in the phagolysosome of mononuclear phagocytic cells of the mammalian host. Promastigotes express a major surface glycoprotein of 63 kDa, referred to as GP63. The expression of GP63 in both Leishmania life stages was studied using rabbit antibodies

against native GP63 as well as rabbit antibodies against recombinant GP63 that was synthesized in an Escherichia coli expression system. Immunofluorescence staining detected GP63 in intracellular amastigotes contained within a macrophage cell line and within freshly isolated lesion amastigotes. Western blot analysis using anti-recombinant GP63 antibodies also demonstrated that amastigotes synthesize GP63 which may undergo differential post-translational processing as compared to promastigote GP63.

10/7/19 (Item 19 from file: 5)  
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10165613 BIOSIS NO.: 199089083504  
LEISHMANIA-TROPICA CHARACTERIZATION OF A LIPOPHOSPHOGLYCAN-LIKE ANTIGEN RECOGNIZED BY SPECIES-SPECIFIC MONOCLONAL ANTIBODIES  
AUTHOR: JAFFE C L (Reprint); PEREZ M L; SARFSTEIN R  
AUTHOR ADDRESS: DEP BIOPHYSICS MACARTHUR CENTER MOLECULAR BIOLOGY TROPICAL DISEASES, WEIZMANN INST SCI, REHOVOT 76100, ISRAEL\*\*ISRAEL  
JOURNAL: Experimental Parasitology 70 (1): p12-24 1990  
ISSN: 0014-4894  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: ENGLISH

ABSTRACT: Species-specific monoclonal antibodies to Leishmania tropica. T11 and T13-15, recognize membranal and secreted antigens. The membrane form of the antigen migrates on sodium dodecyl sulfate-polyacrylamide gels with a diffuse molecular weight from 15 to 50 kDa and can be labeled with palmitic acid, myoinositol, galactose, glucosamine, and inorganic phosphate. Both phosphate and sugarlabeled material were isolated from metabolically labeled promastigotes by affinity chromatography on antibodies coupled to Sepharose 4B. No binding to Ricinus communis agglutinin was observed. This material behaves like lipophosphoglycans from other Leishmania but contains unique species-specific epitopes. It is susceptible to cleavage by phospholipase C and after digestion no longer partition as into the detergent phase following a Triton X-114 extraction. All four monoclonal antibodies appear to recognize carbohydrate epitope on the lipophosphoglycan since

periodate treatment of this material bound to nitrocellulose essentially eliminated antibody binding. In addition, T15 binding could be blocked by 5 mM mannose-6-P04 and fructose-1- or 6-P04, but not by mannose, glucose, fructose, or the additional P04 derivatives examined. The antibodies recognize a similar but not identical epitope, as demonstrated by a competitive radioimmunoassay using 125I-labeled T11, T13, and T15. Expression of surface antigen is elevated during the promastigote stationary phase.

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09175995 BIOSIS NO.: 198886015916  
THE MAJOR SURFACE PROTEIN OF LEISHMANIA PROMASTIGOTES IS A  
FIBRONECTIN-LIKE MOLECULE  
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JOURNAL: European Journal of Immunology 18 (3): p473-476 1988  
ISSN: 0014-2980  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: ENGLISH

ABSTRACT: The major surface glycoprotein of Leishmania chagasi promastigotes showed cross-reactivity with fibronectin (Fn), a large glycoprotein that is a major constituent of the extracellular matrix of most mononuclear cells. Polyclonal and monoclal antibodies against Fn precipitated two molecules of 63-58 kDa from the lysates of both 125I and [35S]methionine-labeled promastigotes. In addition, a monoclonal antibody against a 15-kDa fragment of Fn containing the Arg-Gly-Asp-Ser (RGDS) sequence and several polyclonal monospecific mouse antibodies against a synthetic RGDS peptide also recognized the above two molecules. The attachment of Leishmania promastigotes to mouse peritoneal macrophages in vitro was partially inhibited when promastigotes were treated with F(ab')2 fragment of an anti-Fn IgG. Identical results were obtained by saturating the Fn receptors on macrophages using different peptides containing the RGDS sequence. Moreover, antigen preparations rich in glycoprotein 63 could efficiently promote the attachment and spreading of 3T3 mouse fibroblasts

to surfaces coated with the antigen. These results clearly suggest that the gp63 of *L. chagasi* promastigotes is an Fn-like molecule that

shares certain biological and molecular characteristics with Fn.

10/7/21 (Item 21 from file: 5)  
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09175976 BIOSIS NO.: 198886015897  
IDENTIFICATION OF MONOMERIC AND OLIGOMERIC FORMS OF A MAJOR  
LEISHMANIA-INFANTUM ANTIGEN BY USING MONOCLONAL ANTIBODIES  
AUTHOR: SOTERIADOU K P (Reprint); TZINIA A K; HADZIANTONIOU M G;  
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JOURNAL: Infection and Immunity 56 (5): p1180-1186 1988  
ISSN: 0019-9567  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: ENGLISH

ABSTRACT: Ten monoclonal antibodies (MAbs) produced against isolated *Leishmania infantum* membranes were used as probes of *L. infantum* membrane antigens. Western blots of *L. infantum* membranes, sodium dodecyl sulfate solubilized and heated at 100°C before analysis of sodium dodecyl sulfate-polyacrylamide gel electrophoresis, showed that all 10 MAbs recognized a band at 58 kilodaltons (kDa). However, when solubilized membranes were not heated, 2 of the 10 MAbs recognized, in addition to the 58-kDa band, bands of higher molecular weight. Limited digestion of heated or nonheated membranes showed that both groups of MAbs (i.e., not capable or capable of binding to the high-molecular-weight bands) recognized the same proteolytic digests. Hydrophilic forms of the above proteins, possessing proteolytic activity, were detected and isolated by gel filtration. Protein staining of the isolated monomer analyzed by sodium dodecyl sulfate-polyacrylamide gel electrophoresis, under reducing and heating conditions, revealed incomplete reduction of the 58-kDa protein. The reduced form of the 58-kDa protein migrated at 63 to 65 kDa and was not recognized by the MAbs. These results suggest the existence of a monomeric and an oligomeric form of the 58-kDa antigen. The observed inhibition of *Leishmania* promastigote-macrophage binding caused

by MAbs representative of the two groups (capable of oligomeric and/or monomeric antigen recognition) suggest that the 58-kDa monomer and oligomer play an important role in promastigote-macrophage interaction.

We suggest that the 58-kDa L. infantum antigen is the major surface Leishmania antigen (p63) identified by others.

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08682162 BIOSIS NO.: 198784036311  
CLONING OF A GENE ENCODING THE IMMUNODOMINANT SURFACE ANTIGEN  
OF LEISHMANIA-DONOVANI PROMASTIGOTES  
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JOURNAL: Molecular and Biochemical Parasitology 23 (3): p211-222 1987  
ISSN: 0166-6851  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: ENGLISH

ABSTRACT: This study describes the characterisation of externally oriented surface peptides of both morphological forms of Leishmania donovani, the causative agent of visceral leishmaniasis (kala-azar). Using 125I surface labelling techniques and peptide extraction in the detergents Triton X-100 and Triton X-114, a major iodinated promastigote peptide at 63 kDa or 65 kDa (depending on detergent used) was identified. This peptide was demonstrated to be the immunodominant membrane peptide of L.

donovani and

was strongly recognised by human sera from parasitologically confirmed

cases of kala-azar. This peptide was not demonstrated on the surface of tissue amastigotes, although in vitro translations of poly(A+) RNA from both promastigotes and amastigotes demonstrated that

both forms possessed mRNA that directs the synthesis of a 63 kDa peptide. It is suggested therefore that in amastigotes this peptide may

be a processed antigen. We also report the isolation of a recombinant cDNA clone in the bacteriophage vector  $\lambda$  gt10 which encodes a 63 kDa polypeptide that is recognised by human kala-azar sera. It is proposed that this surface peptide could be used in a specific immunodiagnostic test for leishmaniasis.

10/7/23 (Item 23 from file: 5)  
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08148191 BIOSIS NO.: 198681112082

EXPRESSION AND SIZE HETEROGENEITY OF A 63-KILODALTON MEMBRANE  
GLYCOPROTEIN

DURING GROWTH AND TRANSFORMATION OF LEISHMANIA-MEXICANA-  
AMAZONENSIS

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JOURNAL: Molecular and Biochemical Parasitology 18 (2): p197-210 1986

ISSN: 0166-6851

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: ENGLISH

ABSTRACT: Our previous work by immunoprecipitation with a specific  
monoclonal antibody showed multiple, closely apposed electrophoretic  
bands of a major surface antigen specific to the  
promastigote stage of *Leishmania mexicana amazonensis*.

Here, we analyzed the antigen during growth and transformation of  
this parasite with particular emphasis on the origin of the multiple  
bands. Immunobinding assays revealed the presence of the antigen  
throughout all phases of growth of cloned and uncloned  
promastigotes in

various media for different number of generations. More antigen is  
expressed by promastigotes grown in Medium 199 plus fetal bovine  
serum

than those in serum-supplemented Schneider's medium or a defined  
medium;

however, this is clone-dependent. Purified monoclonal antibody  
coupled to

Affi-Gel 10 gave a high capacity of antigen binding, resolving four  
electrophoretic bands of 60-66 kDa. A 63 kDa membrane  
protein, representing one of the four bands, become predominant  
after

[35S]methionine label and chase. Pretreatment of promastigotes with  
10

µg ml<sup>-1</sup> tunicamycin reduces the antigen to a single band of 54  
kDa. Treatment of the antigen bound to the affinity gel with  
endoglycosidase-H produces similar, but less complete effect. These  
results indicate glycosylation of this antigen with  
asparagine-linked oligosaccharides, which appears to account at  
least in

part for its expression as multiple, closely apposed bands during  
biosynthesis. Binding of fluorescein isothiocyanate-labeled 6H12  
monoclonal IgG or Fab to the promastigotes showed an even  
distribution of

the antigen over the cell surface and its capping upon the  
addition of rabbit anti-mouse IgG. Additional hybridomas prepared  
against

amastigotes yielded monoclonal antibodies which recognized surface antigens common to both stages of the parasite.

10/7/24 (Item 24 from file: 5)  
DIALOG(R)File 5:Biosis Previews(R)  
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07219928 BIOSIS NO.: 198477051839  
RADIO IODINATION AND IDENTIFICATION OF EXTERNALLY DISPOSED MEMBRANE COMPONENTS OF LEISHMANIA-TROPICA  
AUTHOR: GARDINER P R (Reprint); DWYER D M  
AUTHOR ADDRESS: ILRAD, PO BOX 30709, NAIROBI, KENYA\*\*KENYA  
JOURNAL: Molecular and Biochemical Parasitology 8 (4): p283-296 1983  
ISSN: 0166-6851  
DOCUMENT TYPE: Article  
RECORD TYPE: Abstract  
LANGUAGE: ENGLISH

ABSTRACT: Two methods, one involving a lactoperoxidase-glucose oxidase coupled reaction and the other employing the insoluble catalyst 1,3,4,6-tetrachloro-3 $\alpha$ ,6- $\alpha$ -diphenyl-glycoluril (Iodo-Gen), were used to label the surface membrane of promastigotes of L. t. major. Both methods labeled .apprx. 20 proteins or glycoproteins (apparent size range 10-110 kDa [kilodaltons]) in a qualitatively similar manner; however, the lactoperoxidase method labeled 1 additional constituent (260 kDa). By omission of both enzymes, or of Iodo-Gen, by comparison of radioactivity incorporated by particulate and soluble cell fractions, and through the action of proteases on live, labeled promastigotes, the surface-labeling specificity of both procedures was confirmed. Immunoprecipitation of Triton X-100 extracts of labeled cells with rabbit antisera revealed a minimum of 12 (7 major) protein antigens in the homologous system and different but cross-reactive protein species from 2 other isolates of L. tropica. Lectin precipitation of radiolabeled surface components was possible with concanavalin A (but not with other lectins tested) identifying a minimum of 12 glycoproteins. Two of these glycoproteins (120 and 88 kDa) were not recognized by rabbit antiserum.

10/7/25 (Item 1 from file: 34)  
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci  
(c) 2009 The Thomson Corp. All rts. reserv.

11168063 Genuine Article#: 617TU Number of References: 64  
Title: Characterization of Leishmania donovani antigens encapsulated in liposomes that induce protective immunity in BALB/c

mice

Author: Afrin F; Rajesh R; Anam K; Gopinath M; Pal S; Ali N (REPRINT)  
Corporate Source: Indian Inst Chem Biol, Infect Dis Grp, 4 Raja S-C  
Mullick

Rd/Calcutta 700032/W Bengal/India/ (REPRINT); Indian Inst Chem  
Biol, Infect Dis Grp, Calcutta 700032/W Bengal/India/

Journal: INFECTION AND IMMUNITY, 2002, V70, N12 (DEC), P6697-6706

ISSN: 0019-9567 Publication date: 20021200

Publisher: AMER SOC MICROBIOLOGY, 1752 N ST NW, WASHINGTON, DC  
20036-2904

USA

Language: English Document Type: ARTICLE

Abstract: Leishmania donovani promastigote membrane  
antigens (LAG) encapsulated in positively charged liposomes have  
been found to induce very significant levels of protection against  
experimental visceral leishmaniasis. The protectively immunized  
animals

exhibited profound delayed-type hypersensitivity and antibody  
responses. The extent of protection induced by the same antigens,  
however, varied depending on the charge of the vesicles, with

maximum

induction by positively charged liposomes, followed by neutral  
liposomes and last negatively charged liposomes. Characterization

of

LAG and LAG entrapped in liposomes of different charges by

Western blot

analysis revealed the immunodominance of gp63 in all three vaccine  
preparations. The strong reactivity of antigens in a restricted  
antigen profile that included, in addition to gp63, 72-, 52-,  
48-, 45-, 39-, and 20-kDa components in neutral and positively  
charged liposomes contrasted with the reactivity of a greater

number of

LAG components in negatively charged liposomes. Resistance to  
visceral

leishmaniasis appears to depend on the immunity induced by gp63  
and a

few select antigens in association with the right liposomes. A  
striking similarity between the immunogenic profile of partially  
purified soluble antigens and that of LAG in neutral and  
positively charged liposomes suggests the potentiality of these  
antigens in future vaccine studies of L. donovani.

10/7/26 (Item 2 from file: 34)

DIALOG(R)File 34:SciSearch(R) Cited Ref Sci  
(c) 2009 The Thomson Corp. All rts. reserv.

08139365 Genuine Article#: 250FY Number of References: 61

Title: Molecular cloning and characterization of a novel  
repeat-containing

Leishmania major gene, ppg1, that encodes a membrane-associated  
form of proteophosphoglycan with a putative

glycosylphosphatidylinositol anchor  
Author: Ilg T (REPRINT) ; Montgomery J; Stierhof YD; Handman E  
Corporate Source: MAX PLANCK INST BIOL, ABT MEMBRANBIOCHEM, CORRENSSTR  
38/D-72076 TUBINGEN//GERMANY/ (REPRINT); ROYAL MELBOURNE  
HOSP, WALTER &  
ELIZA HALL INST MED RES/MELBOURNE/VIC 3050/AUSTRALIA/  
Journal: JOURNAL OF BIOLOGICAL CHEMISTRY, 1999, V274, N44 (OCT 29), P  
31410-31420  
ISSN: 0021-9258 Publication date: 19991029  
Publisher: AMER SOC BIOCHEMISTRY MOLECULAR BIOLOGY INC, 9650 ROCKVILLE  
PIKE, BETHESDA, MD 20814  
Language: English Document Type: ARTICLE  
Abstract: Leishmania parasites secrete a variety of proteins that are  
modified by phosphoglycan chains structurally similar to those of  
the cell surface glycolipid lipophosphoglycan. These proteins are  
collectively called proteophosphoglycans. We report here the  
cloning and sequencing of a novel Leishmania major proteophosphoglycan  
gene, ppgl. It encodes a large polypeptide of approximately 2300  
amino acids. The N-terminal domain of approximately 70 kDa exhibits 11  
imperfect amino acid repeats that show some homology to  
promastigote surface glycoproteins of the psa2/gp46  
complex. The large central domain apparently consists exclusively  
of approximately 100 repetitive peptides of the sequence  
APSASSSSA(P/S)SSSSS(+/-S). Gene fusion experiments demonstrate  
that these peptide repeats are the targets of phosphoglycosylation in  
Leishmania and that they form extended filamentous structures  
reminiscent of mammalian mucins. The C-terminal domain contains a  
functional glycosylphosphatidylinositol anchor addition signal  
sequence, which confers cell surface localization to a normally  
secreted Leishmania acid phosphatase when fused to its C  
terminus. Antibody binding studies show that the ppgl gene  
product is phosphoglycosylated by phosphoglycan repeats and cap  
oligosaccharides.  
In contrast to previously characterized proteophosphoglycans, the  
ppgl gene product is predominantly membrane-associated and it is  
expressed on the promastigote cell surface. Therefore this  
membrane-bound proteophosphoglycan may be important for direct  
host-parasite interactions.

06844481    Genuine Article#: ZW149    Number of References: 58  
Title: Human and murine immune responses to a novel Leishmania major  
      recombinant protein encoded by members of a multicopy gene family  
Author: Webb JR; CamposNeto A; Ovendale PJ; Martin TI; Stromberg EJ;  
      Badaro

R; Reed SG (REPRINT)

Corporate Source: INFECT DIS RES INST,1124 COLUMBIA ST, SUITE  
      200/SEATTLE//WA/98104 (REPRINT); INFECT DIS RES  
INST,/SEATTLE//WA/98104

; CORIXA CORP,/SEATTLE//WA/98104; UNIV WASHINGTON,DEPT  
      PATHOBIOL/SEATTLE//WA/98195; UNIV FED BAHIA,/SALVADOR/BA/BRAZIL/

Journal: INFECTION AND IMMUNITY, 1998, V66, N7 (JUL), P3279-3289

ISSN: 0019-9567    Publication date: 19980700

Publisher: AMER SOC MICROBIOLOGY, 1325 MASSACHUSETTS AVENUE, NW,  
      WASHINGTON, DC 20005-4171

Language: English    Document Type: ARTICLE

Abstract: Vaccination of BALB/c mice with Leishmania major  
      promastigote culture filtrate proteins plus Corynebacterium  
      parvum confers resistance to infection,vith L. major. To define  
      immunogenic components of this protein mixture, we used sera from  
      vaccinated mice to screen an L. major amastigote cDNA expression  
      library. One of the immunoreactive clones thus obtained encoded a  
novel

      protein of L. major with a molecular mass of 22.1 kDa. The  
      predicted amino acid sequence of this clone exhibited significant  
      homology to eukaryotic thiol-specific-antioxidant (TSA) proteins.  
      Therefore, we have designated this protein L. major TSA protein.  
      Southern blot hybridization analyses indicate that there are

multiple

      copies of the TSA gene in all species of Leishmania analyzed.  
      Northern blot analyses demonstrated that the TSA gene is

constitutively

      expressed in L. major promastigotes and amastigotes. Recombinant

TSA

      protein containing an amino-terminal six-histidine tag was  
expressed in

      Escherichia coli with the pET17b system and was purified to

homogeneity

      by affinity chromatography. Immunization of BALB/c mice with  
      recombinant TSA protein resulted in the development of strong

cellular

      immune responses and conferred protective immune responses against  
      infection with L. major when the protein was combined,vith

interleukin

12. In addition, recombinant TSA protein elicited in vitro  
      proliferative responses from peripheral blood mononuclear cells of  
      human leishmaniasis patients and significant TSA protein-specific  
      antibody titers were detected in sera of both

cutaneous-leishmaniasis

      and visceral-leishmaniasis patients. Together, these data suggest

that

      the TSA protein may be useful as a component of a subunit vaccine

against leishmaniasis.

10/7/28 (Item 4 from file: 34)  
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci  
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06806832 Genuine Article#: ZT861 Number of References: 31  
Title: Leishmania: Amastigotes synthesize conserved secretory acid  
phosphatases during human infection  
Author: Ellis SL; Shakarian AM; Dwyer DM (REPRINT)  
Corporate Source: NIAID, DIV INTRAMURAL RES, PARASIT DIS LAB, CELL BIOL  
SECT, NIH, 9000 ROCKVILLE PI/BETHESDA//MD/20892 (REPRINT);  
NIAID, DIV  
INTRAMURAL RES, PARASIT DIS LAB, CELL BIOL SECT,  
NIH/BETHESDA//MD/20892  
Journal: EXPERIMENTAL PARASITOLOGY, 1998, V89, N2 (JUN), P161-168  
ISSN: 0014-4894 Publication date: 19980600  
Publisher: ACADEMIC PRESS INC JNL-COMP SUBSCRIPTIONS, 525 B ST, STE  
1900,  
SAN DIEGO, CA 92101-4495  
Language: English Document Type: ARTICLE  
Abstract: Leishmania donovani is the major causative agent of Old  
World human visceral leishmaniasis (VL). In vitro, both  
promastigotes  
and axenic amastigotes of L. donovani constitutively secrete  
soluble  
acid phosphatases (SACPs), which contain conserved antigenic  
epitopes. These SACPs are the most abundant and best characterized  
secretory proteins of this parasite. The aim of this study was to  
determine whether this enzyme was produced by intracellular  
amastigotes  
during the course of human infection. To that end, sera from  
acutely  
infected leishmaniasis patients were tested for anti-SACP  
antibodies  
using L. donovani promastigote culture supernatants. Our results  
showed that VL patient sera from different endemic foci  
immunoprecipitated parasite SACP enzyme activity. Further, these  
VL  
patient sera recognized the 110- and 130-kDa SACPs in both  
Western blots and radioimmunoprecipitation assays. Results of  
tunicamycin experiments demonstrated that VL patient anti-SACP  
antibodies were directed against the polypeptide backbone of the  
parasite SACPs. In addition, both radiolabeled L. donovani SACPs  
and  
native enzyme activities were immunoprecipitated by sera from  
patients  
with various forms of cutaneous leishmaniasis. Together these  
studies  
demonstrate that Leishmania amastigotes produce SACPs during the  
course of human infections.

10/7/29 (Item 5 from file: 34)  
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci  
(c) 2009 The Thomson Corp. All rts. reserv.

03622572 Genuine Article#: PT102 Number of References: 48  
Title: IDENTIFICATION OF A 94-KILODALTON ANTIGEN ON LEISHMANIA  
PROMASTIGOTE FORMS AND ITS SPECIFIC RECOGNITION IN HUMAN AND  
CANINE VISCERAL LEISHMANIASIS  
Author: ROLLAND L; ZILBERFARB V; FURTADO A; GENTILINI M  
Corporate Source: 1 RUE LAMARTINE/F-38000 GRENOBLE//FRANCE//; UNIV  
PARIS  
06,PARASITOL LAB/F-75643 PARIS 13//FRANCE//; INST PASTEUR,UNITE  
IMMUNOPARASITOL/F-75724 PARIS//FRANCE//; FDN OSWALDO CRUZ,CTR  
PESQUISAS  
AGGEU MAGALHAES/RECIFE/PE/BRAZIL/  
Journal: PARASITE IMMUNOLOGY, 1994, V16, N11 (NOV), P599-608  
ISSN: 0141-9838  
Language: ENGLISH Document Type: ARTICLE  
Abstract: We have analysed by immunoblotting sera from humans and  
dogs with  
visceral leishmaniasis, from the Old World as well as the New.  
When  
lysates of promastigotes are used as antigens, antibodies against  
a 94 kDa Leishmania component are detected, regardless of  
the age and geographical origin of the patient, the serum antibody  
titre as measured by indirect immunofluorescence, and the number  
of  
arcs in counterimmunoelectrophoresis. Low dilutions of sera from  
patients with Old and New World cutaneous leishmaniasis did not  
react  
with the 94-kDa antigen, whatever the species of  
Leishmania used as antigens. Sera from patients with other  
infections than leishmaniasis, or without infection, are  
negative, even  
at low dilution. Anti-94 kDa antibodies were detected in the sera  
of Leishmania-infected dogs from both the Old and the New World.  
When lysates of leishmania mexicana axenic amastigotes are used  
as antigens, the 94-kDa antigen was little or none  
identified by sera from humans and dogs with visceral  
leishmaniasis,  
and never recognized by control sera. Thus, the specific  
recognition of  
the 94-kDa promastigote antigen in human and canine  
visceral leishmaniasis suggests that this antigen could be a  
potential candidate in the differential immunodiagnosis of the  
disease.

10/7/30 (Item 6 from file: 34)  
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

(c) 2009 The Thomson Corp. All rts. reserv.

03393010    Genuine Article#: PB879    Number of References: 57  
Title: CHARACTERIZATION OF PHOSPHOGLYCAN-CONTAINING SECRETORY  
PRODUCTS OF

LEISHMANIA

Author: ILG T; STIERHOF YD; WIESE M; MCCONVILLE MJ; OVERATH P  
Corporate Source: MAX PLANCK INST BIOL, MEMBRANBIOCHEM ABT, CORRENSSTR  
38/D-72076 TUBINGEN//GERMANY//; MAX PLANCK INST BIOL, MEMBRANBIOCHEM  
ABT/D-72076 TUBINGEN//GERMANY//; UNIV DUNDEE, DEPT BIOCHEM/DUNDEE DD1  
4HN//SCOTLAND/

Journal: PARASITOLOGY, 1994, V108, S, PS63-S71

ISSN: 0031-1820

Language: ENGLISH    Document Type: ARTICLE

Abstract: This article presents an overview on

phosphoglycan-containing

components secreted by the insect and mammalian stages of several  
species of Leishmania, the causative agents of leishmaniasis in  
the Old and New World. Firstly, promastigotes of all three species  
considered, *L. mexicana*, *L. donovani* and *L. major*, shed  
lipophosphoglycan (LPG) into the culture medium possibly by  
release of  
micelles from the cell surface. Like the cell-associated LPG,  
culture supernatant LPG is amphiphilic and composed of a  
lysophosphatidylinositol-phosphosaccharide core connected to  
species-specific phosphosaccharide repeats and oligosaccharide  
caps.

Secondly, all three species release hydrophilic phosphoglycan.

Thirdly,

all three species appear to secrete proteins covalently modified  
by

phosphosaccharide repeats and oligosaccharide caps. In the case of  
promastigotes of *L. mexicana*, these components are organized as  
two

filamentous polymers released from the flagellar pocket: the  
secreted

acid phosphatase (SAP) composed of a 100 kDa phosphoglycoprotein  
and a protein-containing high-molecular-weight-phosphoglycan  
(proteo-HMWPG) and fibrous networks likewise composed of

phosphoglycan

possibly linked to protein. Structural analyses and gene cloning  
suggest that the parasites can covalently modify protein regions  
rich

in serine and threonine residues by the attachment of  
phosphosaccharide

repeats capped by oligosaccharides. We propose that the networks  
formed

in vitro correspond to fibrous material previously demonstrated  
in the

digestive tract of infected sandflies. In the case of *L.*  
*donovani*, the

SAP is also modified by phosphoglycans but contains neither

proteo-HMWPG nor does it aggregate to filaments. Finally, L. mexicana amastigotes release proteo-HMWPG via the flagellar pocket into the parasitophorous vacuole of infected macrophages. This material appears to be released into the tissue of the infected mammal upon rupture of infected macrophages during lesion development. This secretory product may contribute to the pathology of lesion development.

10/7/31 (Item 7 from file: 34)  
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci  
(c) 2009 The Thomson Corp. All rts. reserv.

02868765 Genuine Article#: ML423 Number of References: 26  
Title: CHARACTERIZATION OF A LEISHMANIA ANTIGEN ASSOCIATED WITH  
CYTOPLASMIC VESICLES RESEMBLING ENDOSOMAL-LIKE STRUCTURE  
Author: YAHIAOUI B; LOYENS M; TAIBI A; SCHONECK R; DUBREMETZ JF;  
OUAISSI MA

Corporate Source: INST PASTEUR,CTR IMMUNOL & BIOL  
PARASITAIRE,TRYPANOSOMATIDS RES LAB,INSERM,U167/F-59019  
LILLE//FRANCE/;

INST PASTEUR,CTR IMMUNOL & BIOL PARASITAIRE,TRYPANOSOMATIDS RES  
LAB,INSERM,U167/F-59019 LILLE//FRANCE/; INSERM,U42/F-59045  
LILLE//FRANCE/

Journal: PARASITOLOGY, 1993, V107, DEC (DEC), P497-507  
ISSN: 0031-1820

Language: ENGLISH Document Type: ARTICLE

Abstract: In the present study we have used antibodies to Leishmania major promastigote antigens which were eluted from a glutathione-agarose column (LmGbp) and could identify several parasite components among different Leishmania species by using immunoprecipitation and Western blot techniques. The results also showed that some of LmGbp are present among the molecules released into the culture medium. Moreover, immunofluorescence assays clearly demonstrated that LmGbp are expressed by intracellular amastigotes. The electron micrographs of thawed cryosections of L. major-infected cells revealed that the antigens were associated with the membrane of the phagocytic vacuole. Moreover, the Western blot technique allowed us to identify, using other Leishmania species extracts and anti-LmGbp antibodies, a major polypeptide of an apparent molecular mass of 66 kDa. Immunofluorescence studies suggested that the 66 kDa polypeptide is associated with intracytoplasmic vesicles. Cryosections of Leishmania promastigotes improved the fine

structure preservation of the organelles and enabled a number of features to be seen, particularly the structures considered as vesicles, which appeared as a complex tubulo-vesicular structure resembling mammalian cell endosomes and Leishmania organelles previously named 'megosomes'. Further studies using antibodies against the native 66 kDa protein will be needed to investigate the localization of the protein at the ultrastructural level and to follow its intracellular vesicular traffic.

10/7/32 (Item 8 from file: 34)  
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci  
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02781521 Genuine Article#: MC858 Number of References: 63  
Title: MONOCLONAL-ANTIBODIES DIRECTED AGAINST LEISHMANIA SECRETED  
ACID-PHOSPHATASE AND LIPOPHOSPHOGLYCAN - PARTIAL CHARACTERIZATION  
OF PRIVATE AND PUBLIC EPITOPES  
Author: ILG T; HARBECKE D; WIESE M; OVERATH P  
Corporate Source: MAX PLANCK INST BIOL,CORRENSSTR 38/D-72076  
TUBINGEN//GERMANY/  
Journal: EUROPEAN JOURNAL OF BIOCHEMISTRY, 1993, V217, N2 (OCT 15), P  
603-615  
ISSN: 0014-2956  
Language: ENGLISH Document Type: ARTICLE  
Abstract: Leishmania promastigotes, the stage of the parasite characteristic for the sandfly vector, express an abundant glycoconjugate, called lipophosphoglycan, at their surface. Lipophosphoglycan consists of lysoalkyl-sn-glycerophosphoinositol linked to a phosphosaccharide core conserved in all species, which is connected to P04-6Galbetal,4Manalphal repeats with species-specific substitutions at the Gal residue; the repeats are capped by conserved and species-specific oligosaccharides. Most Leishmania species also secrete an acid phosphatase, which, in Leishmania mexicana, is a filamentous complex composed of a phosphorylated glycoprotein and non-covalently associated proteo-(high-molecular-mass) phosphoglycan.

The secreted acid phosphatase complex was used as an antigen to derive a panel of monoclonal antibodies (mAbs). A total of 25 mAbs (17 novel and 8 previously described) were tested by different techniques for their specificity against lipophosphoglycan and secreted acid phosphatase from several Leishmania species. This comparison

and the modification of the antigens by chemical or enzymic treatments allowed a classification of the mAbs into several groups.

First, from 25 mAbs examined, 22 recognize lipophosphoglycan and the enzyme complex of *L. mexicana*; only three are specific for secreted acid phosphatase. Two of the latter group are also directed against carbohydrate structures, whereas the third mAb recognizes the 100-kDa polypeptide of the complex. The secreted acid-phosphatase-specific class detects antigen in the flagellar pocket of promastigotes while all anti-lipophosphoglycan mAbs bind to the cell surface.

Second, all 15 anti-lipophosphoglycan mAbs investigated in detail appear to be directed against the phosphosaccharide repeats or the cap structure rather than the phosphosaccharide core. Two mAbs recognize terminal cap-structures containing Man $\alpha$ 1,2Man residues. Four antibodies are specific for *L. mexicana* and are probably directed against P04-6[Glc $\beta$ 1,3]Gal $\beta$ 1,4Man $\alpha$ 1 repeats while six mAbs react with the unmodified repeats. Two antibodies specific for *Leishmania major* recognize Gal $\beta$ 1,3-substituted repeats unique for lipophosphoglycan from this species.

Analysis by immunoblotting indicates that the high-molecular-mass proteo-phosphoglycan of *L. mexicana* secreted acid phosphatase carries epitopes for all anti-lipophosphoglycan mAbs suggesting the presence of capped phosphosaccharide repeats while the enzymically active glycoprotein subunit is modified by caps but probably not by repeats. In the case of *Leishmania donovani* secreted acid phosphatase, the enzymically active polypeptide may be directly modified by repeats.

The mAbs are used to characterize changes in lipophosphoglycan structure, which occur in culture during the transition of promastigotes from the logarithmic to the stationary growth phase. Furthermore, testing the mAbs against seven species demonstrates their potential for serotyping *Leishmania*.

10/7/33 (Item 9 from file: 34)  
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci  
(c) 2009 The Thomson Corp. All rts. reserv.

01138614 Genuine Article#: FZ314 Number of References: 30  
Title: BIOCHEMICAL-CHARACTERIZATION OF THE PROTECTIVE MEMBRANE  
GLYCOPROTEIN

GP46/M-2 OF LEISHMANIA-AMAZONENSIS

Author: RIVAS L; KAHL L; MANSON K; MCMAHONPRATT D

Corporate Source: YALE UNIV,SCH MED,DEPT EPIDEMIOLOG & PUBL HLTH,POB  
3333,60

COLL ST/NEW HAVEN//CT/06510; YALE UNIV,SCH MED,DEPT EPIDEMIOLOG &  
PUBL

HLTH,POB 3333,60 COLL ST/NEW HAVEN//CT/06510

Journal: MOLECULAR AND BIOCHEMICAL PARASITOLOGY, 1991, V47, N2,  
P235-243

Language: ENGLISH Document Type: ARTICLE

Abstract: Biochemical features of the immunologically protective,  
membrane

glycoprotein GP46/M-2 of Leishmania amazonensis have been  
investigated. The protein appears to have a single carbohydrate  
side

chain of approximately 3 kDa, representing 7% of the mass of the  
mature GP46/M-2 protein. Experiments removing this carbohydrate  
side

chain from GP46/M-2 indicate that the carbohydrate is not  
involved in  
the epitope recognized by the monoclonal antibody, M-2. As this  
monoclonal antibody recognizes a species-specific epitope, these  
data

suggest that this determinant is defined by the polypeptide  
portion of

the molecule. Studies employing the VSG-lipase as well as  
anti-CRD

antibody clearly indicate that the molecule is anchored to the  
surface membrane of the promastigote via a  
phosphatidylinositol-linked lipid anchor. Neither the  
carbohydrate

side chain nor the lipid anchor appear to be responsible for the  
apparent refractoriness of this protein to protease digestion,  
suggesting that properties of the polypeptide itself may be  
responsible. These data are discussed in terms of recent

DNA-derived  
protein sequence of the GP46/M-2.

10/7/34 (Item 1 from file: 50)  
DIALOG(R)File 50:CAB Abstracts  
(c) 2009 CAB International. All rts. reserv.

0008538736 CAB Accession Number: 20033189694

Immune complex antigens as a tool in serodiagnosis of kala-azar.

Tapati Chakraborti; Dwijen Sarkar; Ghosh, D. K.  
Author email address: t\_chakraborti@yahoo.com  
Department of Biochemistry and Biophysics, University of  
Kalyani,  
Kalyani 741 235, West Bengal, India.  
Molecular and Cellular Biochemistry volume 253 (1/2): p.191-198  
Publication Year: 2003  
ISSN: 0300-8177  
Digital Object Identifier: 10.1023/A:1026095328695  
Additional Title: Special issue: recent advances in molecular  
physiology  
Publisher: Kluwer Academic Publishers Dordrecht, Netherlands  
Language: English  
Record Type: Abstract  
Document Type: Journal article

The 63 kDa surface antigen of Leishmania promastigotes is one of the most important virulent factors in establishing the host parasite relationship. This glycoprotein is revealed by surface iodination study as well as by metabolic labeling and immunoblot methods. In search of this specific antigen for serodiagnosis, immune complexes (ICs) were isolated from kala-azar patient sera and analysed by SDS-PAGE and Western immunoblotting. The immunoblot of kala-azar IC with patient sera, anti-promastigote sera and anti gp63 sera detected the major antigen of 55 kDa. This recognition suggests that 55 kDa antigen and gp63 have common antigenic epitope(s). Normal IC did not react with anti gp63 sera indicating absence of this antigen in normal IC. To confirm the parasitic origin of the 55 kDa antigen of kala-azar IC, in vitro IC was formed with parasite antigen and acid dissociated kala-azar IC antibody. This indicated the antigenic similarity of the 55 kDa antigen and gp63 antigen of the parasite. This also suggested that the former antigen may have been processed from gp63. In summary, identification of parasite antigen (55 kDa) in IC of kala-azar patients' sera may be useful in developing a serodiagnostic assay of visceral leishmaniasis. Several other antigens are visualized in kala-azar IC when developed with patient sera. But specificity and efficacy of these antigens have not yet been evaluated in serodiagnosis of the disease.

42 reference

10/7/35 (Item 2 from file: 50)  
DIALOG(R)File 50:CAB Abstracts  
(c) 2009 CAB International. All rts. reserv.

0008526162 CAB Accession Number: 20033177260

Extracellular release of the surface metalloprotease, gp63, from Leishmania and insect trypanosomatids.

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Cell Biology Section, Div. of Intramural Research, NIAID, NIH, Bethesda, MD 20892, USA.

Parasitology Research volume 91 (3): p.229-237

Publication Year: 2003

ISSN: 0932-0113

Digital Object Identifier: 10.1007/s00436-003-0960-0

Publisher: Springer-Verlag Berlin, Germany

Language: English

Record Type: Abstract

Document Type: Journal article

Protease activity was found in spent culture medium collected from Leishmania donovani, L. mexicana, L. major, as well as the insect trypanosomatids, Crithidia luciliae and Leptomonas seymouri.

Released

protease activity increased linearly over time and was correlated to

promastigote density. In SDS-PAGE, zymogram gels showed that the protease's molecular weight ranged from 43-100 kDa. Spent culture medium proteases were blocked by the metallo-protease inhibitors,

1,10-phenanthroline and Z-Tyr-Leu-NHOH, but not by bestatin, leupeptin,

ABESF, pepstatin A, E-64 or aprotinin. Monoclonal and/or polyclonal

antibodies to the leishmanial gp63 reacted with the released Crithidia,

Leptomonas, L. major and L. donovani proteases. Cell surface biotinylation and immune precipitation using gp63-specific antibodies

showed that >34% of the released protease originated from the

surface. Antibodies against the Trypanosoma brucei variable surface glycoprotein cross-reactive determinant (CRD) did not recognize this activity, suggesting that the gp63 is not cleaved from the

cell surface by a parasite phospholipase, but is released by an alternative mechanism.

48 reference

10/7/36 (Item 3 from file: 50)

DIALOG(R)File 50:CAB Abstracts

(c) 2009 CAB International. All rts. reserv.

0008432384 CAB Accession Number: 20033079688

Characterization of Leishmania donovani antigens encapsulated in liposomes that induce protective immunity in BALB/c mice.

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Infection and Immunity volume 70 (12): p.6697-6706

Publication Year: 2002

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Digital Object Identifier: 10.1128/IAI.70.12.6697-6706.2002

Publisher: American Society for Microbiology (ASM) Washington, USA

Language: English

Record Type: Abstract

Document Type: Journal article

Leishmania donovani promastigote membrane antigens (LAG) encapsulated in positively charged liposomes have been found to induce very significant levels of protection against experimental visceral leishmaniasis. The protectively immunized animals exhibited profound delayed-type hypersensitivity and antibody responses. The extent of protection induced by the same antigens, however, varied depending on the charge of the vesicles, with maximum induction by positively charged liposomes, followed by neutral liposomes and last negatively charged liposomes. Characterization of LAG and LAG entrapped in liposomes of different charges by Western blot analysis revealed the immunodominance of gp63 in all three vaccine preparations. The strong reactivity of antigens in a restricted antigen profile that included, in addition to gp63, 72-, 52-, 48-, 45-, 39-, and 20-kDa components in neutral and positively charged liposomes contrasted with the reactivity of a greater number of LAG components in negatively charged liposomes. Resistance to visceral leishmaniasis appears to depend on the immunity induced by gp63 and a few select antigens in association with the right liposomes. A striking similarity between the immunogenic profile of partially purified soluble antigens and that of LAG in neutral and

positively charged liposomes suggests the potentiality of these antigens in future vaccine studies of *L. donovani* .  
64 reference

10/7/37 (Item 4 from file: 50)  
DIALOG(R)File 50:CAB Abstracts  
(c) 2009 CAB International. All rts. reserv.

0008119239 CAB Accession Number: 20013022440

Regulation of GP63 mRNA stability in promastigotes of virulent and attenuated *Leishmania chagasi* .

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52242, USA.

Molecular and Biochemical Parasitology volume 112 (1): p.51-59  
Publication Year: 2001  
ISSN: 0166-6851  
Digital Object Identifier: 10.1016/S0166-6851(00)00346-7  
Publisher: Elsevier Science Ltd Oxford, UK  
Language: English  
Record Type: Abstract  
Document Type: Journal article

GP63 is a 63-kDa glycoprotein that is abundantly expressed on the surface of all *Leishmania* species and is involved in several steps of promastigote infection of host cells. *Leishmania chagasi* [*Leishmania infantum chagasi*] has at least 18 haploid msp (major surface protease) genes encoding GP63 that are divided into three classes, msp S, msp L or msp C, according to their unique 3prime UTR sequences and differential expression. All three msp classes are constitutively transcribed during virulent promastigote growth in vitro, although msp L mRNA is most abundant during logarithmic phase and msp S mRNA predominates in stationary phase. Thus, the steady state levels of the msp L and msp S mRNAs are post-transcriptionally regulated. Using Actinomycin D to arrest transcription, we found that in virulent promastigotes the half-life ( $t_{SUB\ 1/2}$ ) of msp L mRNA is coordinately modulated with growth phase, decreasing from a mean of 84 min. during early logarithmic growth to a mean of 17 min. at a stage intermediate between logarithmic and stationary phase. However, in attenuated

promastigotes, the  $t_{SUB\ 1/2}$  of msp L RNA remains the same throughout parasite growth. In contrast to msp L RNA, the  $t_{SUB\ 1/2}$  of msp S and msp C RNA is constant throughout all growth phases of both virulent and attenuated promastigote growth. The presence of the translation inhibitor cycloheximide increases the  $t_{SUB\ 1/2}$  of msp L RNA 4-6-fold in both virulent and attenuated promastigotes at all growth phases. These results indicate that the  $t_{SUB\ 1/2}$  of msp L RNA is maintained by at least two distinct mechanisms - one activated during growth to stationary phase and the other dependent on a labile negative regulatory protein factor(s).

36 reference

10/7/38 (Item 5 from file: 50)  
DIALOG(R)File 50:CAB Abstracts  
(c) 2009 CAB International. All rts. reserv.

0007947182 CAB Accession Number: 20000809086

Expression and immunological identification of recombinant GP63 of

*Leishmania major*.

Yan Lei; Zuo XinPin; Hou YanYan ( et al. )

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Xinjiang 830002, China.

Endemic Diseases Bulletin volume 15 (2): p.13-14, 16

Publication Year: 2000

ISSN: 1000-3711

Language: Chinese Summary Language: English

Record Type: Abstract

Document Type: Journal article

Recombinant *Leishmania major* surface glycoprotein GP63 was expressed in *Escherichia coli* XL1-Blue transformed with the recombinant plasmid PAS 26, which was constructed from the pBluescript M13 plasmid and the cloned GP63 gene from *L. major*. The molecular weight of the expression product was 55 kDa after dissociation from beta-galactosidase. A low titre of cross reaction was seen between immune sera against recombinant GP63 and beta-galactosidase from bacteria. A high titre in ELISA was obtained using recombinant GP63 with immune sera

against Leishmania promastigote antigen and with sera from kala-azar patients. It is concluded that this recombinant GP63 could be useful in serological diagnosis of kala-azar and research on immunization against the disease.  
2 reference

10/7/39 (Item 6 from file: 50)  
DIALOG(R)File 50:CAB Abstracts  
(c) 2009 CAB International. All rts. reserv.

0007522001 CAB Accession Number: 19980802636

Cloning of the gp63 surface protease of Leishmania infantum  
. Differential post-translational modifications correlated with different infective forms.

Gonzalez-Aseguinolaza, G.; Almazan, F.; Rodriguez, J. F.; Marquet, A.; Larraga, V.  
Centro de Investigaciones Biologicas, CSIC, Velazquez 144, Madrid, Spain.

Biochimica et Biophysica Acta, Molecular Basis of Disease volume 1361 (1

): p.92-102

Publication Year: 1997

ISSN: 0925-4439

Language: English

Record Type: Abstract

Document Type: Journal article

The gp63 gene from Leishmania infantum [ L. infantum infantum ] PB75 was cloned and characterized. The sequence analysis of the gene indicated the existence of a high degree of conservation with the other

Old World species L. major and L. donovani . The similarity was lower with

New World species with the exception of L. chagasi [ L. infantum chagasi ]

which showed a strikingly high percentage of identity (99-100%). In L.

infantum infantum the gp63 gene expressed 2 polypeptides of 58 and 60

kDa , respectively, which showed a similar proteolytic activity. The 60 kDa polypeptide was expressed during the whole life cycle of the promastigote form of the parasite with a moderate increase at the stationary phase of growth, whereas the 58 kDa product, although slightly present in the logarithmic phase, notably increased its

expression during the highly infectious stationary phase. RNA analysis showed that the presence in *L. infantum chagasi* of these 2 polypeptides correlated with 2 RNA molecules and with the degree of parasite infectivity, whereas in the case of *L. infantum infantum* a single 3 kb messenger RNA was detected through the whole promastigote life cycle. The data indicated that in *L. infantum infantum*, the differences in gene expression of the gp63 protease family according to parasite phase of growth seems to be due to a differential pattern of glycosylation of the polypeptides which correlates with the different infective forms of the promastigote form of the parasite. Nucleotide sequence data have been submitted to GenBank under accession number U48798.  
46 reference

10/7/40 (Item 7 from file: 50)  
DIALOG(R)File 50:CAB Abstracts  
(c) 2009 CAB International. All rts. reserv.

0006222555 CAB Accession Number: 19902070150  
Identification of cytoplasmic soluble antigens related to the major surface antigens of *Leishmania braziliensis braziliensis* and *L. donovani chagasi*.  
Kutner, S.; Pellerin, P.; Breniere, S. F.  
Dept Cell Biol., Faculty Scis, University Cent. Venezuela, PO Box 47069, Los Chaguaramos, Caracas 1041, Venezuela.  
Parasitology Research volume 76 (3): p.185-191  
Publication Year: 1990  
ISSN: 0044-3255  
Language: English  
Record Type: Abstract  
Document Type: Journal article

This study describes the identification of aqueous-soluble antigens in *Leishmania* promastigotes immunologically and biochemically closely related to the major surface antigen. Proteins from surface-iodinated *L. braziliensis braziliensis* and *L. donovani chagasi* promastigotes, extracted and separated by partitioning in the detergent Triton X-114, were analyzed. Immunoblotting of the extracted proteins, using homologous antisera, showed recognition of a 72-kDa labeled, amphiphilic antigen of *L. b. braziliensis* and a 65-

kDa surface antigen of *L. d. chagasi*. The respective homologous sera also recognized non-labeled hydrophilic antigens, similar in their apparent molecular weights to the major surface antigens. The amphiphilic and hydrophilic antigens of each species were found to share common antigenic determinants, inasmuch as monospecific antibodies that recognized the amphiphilic protein reacted

with the hydrophilic antigen. Structural homology was also obtained in the peptide-digestion profiles of the amphiphilic and the respective

hydrophilic major antigens. Zymogram assay showed that both amphiphilic and hydrophilic fractions displayed proteolytic activity that

could be directly attributed to the major *L. b. braziliensis* and *L. d.*

*chagasi* antigens. The hydrophilic antigens found in this study are probably not hydrolytic products of the surface antigens and occur in large quantities in the promastigote cytosol.

This study describes the identification of aqueous-soluble

antigens in *Leishmania* promastigotes immunologically and biochemically closely related to the major surface antigen. Proteins from surface-iodinated *L. braziliensis braziliensis* and *L. donovani chagasi* promastigotes, extracted and separated by partitioning in

the detergent Triton X-114, were analyzed. Immunoblotting of the extracted

proteins, using homologous antisera, showed recognition of a 72-kDa labeled, amphiphilic antigen of *L. b. braziliensis* and a 65-kDa surface antigen of *L. d. chagasi*. The respective

homologous sera also recognized non-labeled hydrophilic antigens, similar in their apparent molecular weights to the major surface antigens. The amphiphilic and hydrophilic antigens of each species were found to share common antigenic determinants, inasmuch as monospecific antibodies that recognized the amphiphilic protein reacted

with the hydrophilic antigen. Structural homology was also obtained in the peptide-digestion profiles of the amphiphilic and the respective

hydrophilic major antigens. Zymogram assay showed that both amphiphilic and hydrophilic fractions displayed proteolytic activity that

could be directly attributed to the major *L. b. braziliensis* and *L. d.*

*chagasi* antigens. The hydrophilic antigens found in this study are probably not hydrolytic products of the surface antigens and occur in large quantities in the promastigote cytosol.

24 reference

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0003195596 SUPPLIER NUMBER: 1999195672

Interaction of Leishmania gp63 with cellular receptors for fibronectin

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1999, United States

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DOCUMENT TYPE: Article

LANGUAGES: English SUMMARY LANGUAGES: English

NO. OF REFERENCES: 37

The most abundant protein on the surface of the promastigote form of the protozoan parasites *Leishmania* spp. is a 63-kDa molecule, designated gp63 or leishmanolysin. Because gp63 has been shown to possess fibronectin-like properties, we examined the interaction of gp63 with the cellular receptors for fibronectin. We measured the direct binding of *Leishmania* to human macrophages or to transfected mammalian cells expressing human fibronectin receptors. *Leishmania* expressing gp63 exhibited modest but reproducible adhesion to human macrophages and to transfected CHO cells expressing alpha4/beta1 fibronectin receptors. In both cases, this interaction depended on gp63 but occurred independently of the SRYD sequence of gp63, because parasites expressing gp63 with a mutated SRYD sequence bound to macrophages and alpha4/beta1 receptor-expressing cells as well as did wild-type parasites. The contribution of gp63 to parasite adhesion was more pronounced when the assays were performed in the presence of complement, suggesting that the receptors for complement and fibronectin may cooperate to mediate the efficient adhesion of parasites to macrophages. The interaction of gp63 with fibronectin receptors may also play an important role in parasite internalization by macrophages.

Erythrocytes to which gp63 was cross-linked were efficiently phagocytized by macrophages, whereas control erythrocytes opsonized with complement alone bound to macrophages but remained peripherally attached to the outside of the cell. Similarly, parasites expressing wild-type gp63 were rapidly and efficiently phagocytized by resting macrophages, whereas parasites lacking gp63 were internalized more slowly. This rapid internalization of gp63-expressing parasites was dependent on the beta1 integrins, because pretreatment of macrophages with monoclonal antibodies to the beta1 integrins decreased the internalization of gp63-expressing parasites. These observations indicate that complement receptors are the primary mediators of parasite adhesion; however, maximal parasite adhesion and internalization may require the participation of the beta1 integrins, which recognize fibronectin-like molecules such as gp63 on the surface of the parasite.

10/7/42 (Item 1 from file: 72)  
DIALOG(R)File 72:EMBASE  
(c) 2009 Elsevier B.V. All rts. reserv.

0079685472 EMBASE No: 2003394515  
Identification of a gene in Leishmania infantum encoding a protein that contains a SP-RING/MIZ zinc finger domain  
Garcia-Estrada C.; Reguera R.M.; Villa H.; Requena J.M.; Muller S.; Perez-Pertejo Y.; Balana-Fouce R.; Ordonez D.  
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Biochimica et Biophysica Acta - Gene Structure and Expression ( Biochim. Biophys. Acta Gene Struct. Expr. ) (Netherlands) October 1, 2003, 1629/1-3 (44-52)  
CODEN: BBGSD ISSN: 0167-4781  
DOI: 10.1016/j.bbaexp.2003.07.001  
DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract  
LANGUAGE: English SUMMARY LANGUAGE: English  
NUMBER OF REFERENCES: 34  
The SP-RING or Miz zinc finger domain that is related to the classical

RING-finger motif, defines a class of proteins that can act as E3-like factors in the pathway of small ubiquitin-related modifier (SUMO) conjugation. This family includes the mammalian protein inhibitor of activated STAT (PIAS) proteins and related proteins from lower eukaryotes.

Here we report the existence of a gene in *Leishmania infantum*, present as two identical copies placed upstream of each MAT2 gene copy, and transcribed as a single (similar) 2.2 kb mRNA both in the logarithmic and stationary phases of the promastigote stage. This gene encodes a 47 kDa protein that has been named LORIEN. LORIEN is circumscribed to the cell periphery and it is antigenic during *L. infantum* infection of dogs and hamsters. Strikingly, this novel protein contains a highly conserved SP-RING/Miz zinc finger domain, raising the possibility that a SUMO or ubiquitin-like system may exist in this microorganism. (c) 2003 Elsevier B.V. All rights reserved.

10/7/43 (Item 2 from file: 72)  
DIALOG(R)File 72:EMBASE  
(c) 2009 Elsevier B.V. All rts. reserv.

0075659276 EMBASE No: 1994087105

*Leishmania* major parasites share an epitope with the murine CD3-T cell receptor complex

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European Journal of Immunology ( EUR. J. IMMUNOL. ) (Germany)  
March 1,  
1994, 24/3 (503-507)

CODEN: EJIMA ISSN: 0014-2980

DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract

LANGUAGE: English SUMMARY LANGUAGE: English

NUMBER OF REFERENCES: 20

After immunization of BALB/c mice with a low molecular mass fraction (FrD;  $\leq$  31 kDa) isolated from a soluble extract of *Leishmania* major promastigotes, a panel of monoclonal antibodies (mAb) was obtained.

One of these antibodies (mAb 9C) recognized a cytosol-associated antigen from *L. major* of approximately 21 kDa as shown by

Western blot and immunoprecipitation. In addition, mAb 9C reacted with surface structures of murine splenic T cells and T cell clones. Reactivity was confined to murine cells, but was not strain restricted.

Immunoprecipitation studies and surface-labeling experiments with CD4 SUP + T cell clones and the T cell receptor (TCR) SUP -CD3 SUP - T cell line TG40 transfected with V alpha/beta chains from human TCR and concomitant co-expression of murine CD3 suggested that mAb 9C binds to an epitope located within the murine CD3-TCR complex. In addition, mAb 9C induced strong T cell proliferation. We conclude that L. major parasites share an epitope with the murine CD3-TCR complex which is functionally important for T cell activation.

10/7/44 (Item 1 from file: 73)  
DIALOG(R)File 73:EMBASE  
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0074694298 EMBASE No: 1991199109  
Metacyclogenesis of Leishmania spp: Species-specific in vitro transformation, complement resistance, and cell surface carbohydrate and protein profiles

Bandyopadhyay P.; Ghosh D.K.; De A.; Ghosh K.N.; Chaudhuri P.P.; Das P.; Bhattacharya A.

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Journal of Parasitology ( J. PARASITOL. ) (United States) July 24, 1991, 77/3 (411-416)

CODEN: JOPAA ISSN: 0022-3395

DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract

LANGUAGE: English SUMMARY LANGUAGE: English

Metacyclic (stationary) and logarithmic (log) forms of promastigotes of Leishmania donovani and Leishmania major were characterized in several ways. The highly active metacyclic forms were larger with more protein and less carbohydrate. The flagellum increased in length 2.4 times in L. major as compared to 1.8 times in L. donovani. Resistance to

complement-mediated lysis by normal human serum of in vitro grown *Leishmania* promastigotes was related to the species, the growth phase in culture, and also the temperature. Metacyclic forms of both species had a much increased resistance to killing by normal serum at different temperatures. Differences in membrane-exposed carbohydrates were detected by fluorescein-conjugated lectins. Peanut agglutinin and Ulex agglutinin I differentiated log and stationary phase promastigotes of *L. major*. Higher amounts of acid phosphatase were demonstrated in the metacyclic phase. Differences in polypeptides were detected by sodium dodecyl sulfate-polyacrylamide gel electrophoresis. Two polypeptides of approximately 51 and 114 kDa were found exclusively in metacyclic promastigotes of both species, whereas 38- and 23-kDa polypeptides were lost or reduced during transformation from log to metacyclic phase promastigotes of *L. donovani*. In addition, a 75-kDa polypeptide was expressed only in metacyclic promastigotes of *L. major*.

10/7/45 (Item 2 from file: 73)  
DIALOG(R)File 73:EMBASE  
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0074667156 EMBASE No: 1991171967  
Localization of the plasma membrane and mitochondrial H SUP  
+-ATPases in  
*Leishmania donovani* promastigotes  
Liveanu V.; Webster P.; Silberstein D.  
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European Journal of Cell Biology ( EUR. J. CELL BIOL. ) (Germany)  
July  
1, 1991, 54/1 (95-101)  
CODEN: EJCB D ISSN: 0171-9335  
DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract  
LANGUAGE: English SUMMARY LANGUAGE: English

Immunochemical methods were used to characterize the  
proton-translocating  
ATPases (H SUP +-ATPases) of the plasma membrane and mitochondrion of  
*Leishmania donovani* promastigotes. Antisera directed against the  
plasma membrane H SUP +-ATPase of *Saccharomyces cerevisiae* reacted  
with a  
66 kDa membrane protein of *L. donovani* promastigotes. By  
immunocytochemistry, the antiserum was shown to label the cell and

flagellar surface of promastigotes as well as the Golgi apparatus and the membrane of intracellular organelles. The target antigen was shown to possess ATPase activity resembling the leishmanial H SUP +-ATPase activity. Antisera raised against the beta-subunit of the F SUB OF SUB 1-ATPase of *Escherichia coli* reacted with a 56 kDa protein in *L. donovani* promastigotes. Ultrastructurally, the anti-beta-subunit antibody was exclusively associated with the mitochondrion in these cells. This antiserum immunoprecipitates ATP hydrolytic activity typical of the F SUB 1 beta-subunit activity of the mitochondria of higher eukaryotes.

10/7/46 (Item 3 from file: 73)  
DIALOG(R)File 73:EMBASE  
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0074598336 EMBASE No: 1991103836  
Antigenic specificity of the 72-kilodalton major surface glycoprotein of *Leishmania braziliensis braziliensis*  
Kutner S.; Pellerin P.; Breniere S.F.; Desjeux P.; Dedet J.P.  
Inst. de Biologia de Altura, Embajada de Francia, Casilla 717, La Paz,  
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Journal of Clinical Microbiology ( J. CLIN. MICROBIOL. ) (United States)  
April 11, 1991, 29/3 (595-599)  
CODEN: JCMID ISSN: 0095-1137  
DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract  
LANGUAGE: English SUMMARY LANGUAGE: English

We examined the expression and the antigenicity of the major surface polypeptides of *Leishmania braziliensis braziliensis* and *Leishmania donovani chagasi*, parasites which commonly coexist in the same endemic areas of Bolivia. Sodium dodecyl sulfate-polyacrylamide gel electrophoresis protein profiles from surface-iodinated promastigotes showed the presence of a unique iodinated polypeptide of 72 kDa on the *L. b. braziliensis* surface and of two major components of 65 and 50 kDa exposed at the surface of *L. d. chagasi*. Comparison of the peptide digestion profiles of the major iodinated polypeptides of both strains showed no similarity between the maps of the 72- and the 65-kDa polypeptides of *L. b. braziliensis* and *L. d. chagasi*, respectively. Immunoprecipitation of surface-labeled *L. b. braziliensis* Nonidet P-40 extracts with 35 serum specimens obtained

from Bolivian patients with cutaneous and mucocutaneous leishmaniasis showed that all serum specimens recognized predominantly the 72 kDa antigen and high-molecular-mass proteins in some cases. The recognition patterns were independent of the geographical origin of the patient, the type of lesion, and the serum antibody titer. Serum specimens from children with visceral leishmaniasis did not precipitate the L. b. braziliensis 72-kDa antigen. Hamster hyperimmune serum against L. b. braziliensis also recognized the 72-kDa surface antigen. However, this recognition was inhibited in the presence of the homologous nonlabeled antigen but not in the presence of heterologous (L. d. chagasi and Trypanosoma cruzi) antigens. The specific recognition of the 72-kDa surface antigen in both natural and experimental L. b. braziliensis infections suggests that this antigen could be a good candidate for use in the differential immunodiagnosis and prognosis of the disease.

10/7/47 (Item 4 from file: 73)  
DIALOG(R)File 73:EMBASE  
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0074554918 EMBASE No: 1991060418  
Evidence of transferrin binding sites on the surface of  
Leishmania promastigotes  
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21,  
Greece

Journal of Biological Chemistry ( J. BIOL. CHEM. ) (United States)  
December 1, 1990, 265/36 (22380-22385)  
CODEN: JBCHA ISSN: 0021-9258  
DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract  
LANGUAGE: English SUMMARY LANGUAGE: English

A glycoprotein of 78,000 molecular mass (78 kDa), associated with the membrane of Leishmania infantum promastigotes, was identified and immunopurified by monoclonal antibody (mAb) LD9 produced against isolated membrane preparations. mAb LD9 was subsequently found to bind to human transferrin, also of 78 kDa. Binding of LD9 to transferrin was completely abolished when the mAb was preabsorbed by Leishmania membranes, thereby indicating that the 78-kDa Leishmania membrane-associated glycoprotein and transferrin have common

antigenic epitope(s). The 78-kDa Leishmania membrane-associated protein was released in soluble nonaggregated form by mild treatment with acetic acid saline. Anti-transferrin polyclonal antibodies, recognized both the membrane-associated and the soluble form of the 78-kDa glycoprotein. The 78-kDa soluble form was characterized further as an iron-containing protein. The above data combined with iron uptake by promastigotes as demonstrated by the Prussian blue reaction indicate that the 78-kDa Leishmania membrane-associated glycoprotein is transferrin. The binding of SUP 125I-human transferrin to Leishmania-purified membrane preparations was then investigated. The results indicate the presence of a high affinity saturable binding site [ $K(d) = 2.2 \cdot 10^{-8}$  M] that is specific for transferrin. We suggest that the 78-kDa glycoprotein recognized by mAb LD9 is transferrin that binds to the surface of Leishmania promastigotes via a transferrin receptor.

10/7/48 (Item 5 from file: 73)  
DIALOG(R)File 73:EMBASE  
(c) 2009 Elsevier B.V. All rts. reserv.

0074263395 EMBASE No: 1990157408  
Biochemical evidence of the antigenic cell surface  
heterogeneity of *Leishmania mexicana*  
Manenti S.; Kutner S.; Rascon A.; Hernandez A.G.  
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Parasitology Research ( PARASITOL. RES. ) (Germany) June 20, 1990,  
76/4  
(301-305)  
CODEN: PARRE ISSN: 0044-3255  
DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract  
LANGUAGE: English SUMMARY LANGUAGE: English

In the present study, an enzymatical and structural analysis of *Leishmania mexicana* cell-surface components was carried out, demonstrating that protease and acid phosphatase activities were present at the *L. mexicana* cell surface. These findings correlate with the expression of the main components detected on the surface of *L. mexicana* promastigotes: the 50-kDa component is responsible for the

acid phosphatase activity, whereas glycoprotein 65 (gp65) was characterized as the structural polypeptide of the surface protease. Furthermore, the 50- and 65-kDa antigens were found to be structurally different, inasmuch as no homology was observed in their peptide digestion profiles. The results presented in this communication confirm heterogeneity in the expression of the surface components of *L. mexicana* promastigotes at both the structural and the biochemical level.

10/7/49 (Item 6 from file: 73)  
DIALOG(R)File 73:EMBASE  
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0073714390 EMBASE No: 1988175283

The major concanavalin A-binding surface glycoprotein of *Leishmania donovani* chagasi promastigotes is involved in attachment to human macrophages

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Journal of Immunology ( J. IMMUNOL. ) (United States) August 15, 1988,  
141/1 (265-272)

CODEN: JOIMA ISSN: 0022-1767

DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract

LANGUAGE: English SUMMARY LANGUAGE: English

*Leishmania donovani*, the protozoan causing visceral leishmaniasis, is an obligate intracellular parasite of mammalian macrophages. Considerable evidence has suggested that the ingestion of *L. donovani* promastigotes by macrophages occurs via receptors on the surface of the phagocyte. During this study, a glycoconjugate that may be involved in the receptor-mediated ingestion of *L. donovani* chagasi promastigotes was isolated from the parasite membrane. Octyl glucoside-soluble extracts of promastigote membranes contained a predominant doublet migrating at 60 kDa, seen by SDS-PAGE. The 60-kDa molecule was the major externally disposed promastigote surface protein labeled by SUP 125I, and it was the major Con A-binding protein on *L. donovani* chagasi, as

determined by Con A binding to parasite proteins transferred to nitrocellulose. Attachment of promastigotes to human monocyte-derived macrophages was inhibited by varying concentrations of the membrane extract containing both proteins, and adsorption of extracts on Con A-Sepharose resulted in both removal of the 60,000 M(r) glycoprotein and loss of the ability of extracts to inhibit promastigote attachment to human macrophages. After further purification of the 60-kDa glycoprotein by gel filtration, its inhibitory activity increased 45-fold over the unpurified membrane extract. Examination of Con A blots of stationary phase promastigotes isolated from an infected hamster revealed a marked loss in the major Con A-binding glycoprotein over 4 mo in in vitro culture after isolation from the rodent host, corresponding to a loss in infectivity of the promastigotes for hamsters. The results suggest that the major Con A-binding surface glycoprotein from *L. donovani* chagasi promastigotes is important in attachment to human macrophages, and may be a factor in parasite virulence for a mammalian host.

10/7/50 (Item 1 from file: 144)  
DIALOG(R)File 144:Pascal  
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08889818 PASCAL No.: 90-0057784  
L'infectivite des promastigotes de Leishmania est associee a l'expression de leurs antigenes de surface  
(Infectivity of Leishmania promastigotes is associated with surface antigenic expression)  
RIZVI Farrukh; CAPRON Andre, Dir the  
Univ.: Lille 1 Degree: Th. doct. : Immunol.  
1989; 1989 145 p.  
Availability: CNRS-T Bordereau  
No. of Refs.: 165 ref.  
Document Type: T (Thesis) ; M (Monographic)  
Country of Publication: France  
Language: French  
Caracterisation des molecules de surface de Leishmania chagasi impliquees dans l'attachement du promastigote au macrophage.  
L'infectivite des promastigotes est associee a l'expression d'une glycoproteine de surface de masse moleculaire 63 kDa (gp 63). mise en evidence d'une communaute antigenique entre la gp63 et la fibronectine. Il semble que le parasite puisse adherer au macrophage grace aux recepteurs fibronectines de celui-ci

10/7/51 (Item 1 from file: 154)  
DIALOG(R)File 154:MEDLINE(R)  
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14317997 PMID: 11432790

Role of 67 kDa cell surface laminin binding protein of  
Leishmania donovani in pathogenesis.

Bandyopadhyay K; Karmakar S; Ghosh A; Das P K

Molecular Cell Biology Laboratory, Indian Institute of Chemical  
Biology,

Raja S.C. Mullick Road, Jadavpur, Calcutta 700 032, India.

Journal of biochemistry (Japan) Jul 2001, 130 (1) p141-8,  
ISSN

0021-924X--Print Journal Code: 0376600

Publishing Model Print

Document type: Journal Article; Research Support, Non-U.S. Gov't

Languages: ENGLISH

Main Citation Owner: NLM

Record type: MEDLINE; Completed

The role that interaction with laminin may play in Leishmania  
donovani infection was investigated. Binding of

(125)I-radiolabeled

laminin, in a liquid-phase assay, by the parasite was rapid,  
saturable,

specific, reversible, and of high affinity. Using a Western  
blotting

procedure, a 67 kDa laminin-binding protein (LBP) was identified from  
the membrane of both the promastigote and amastigote forms of L.

donovani. Subsequently, the protein was purified by  
affinity

chromatography. Immunofluorescence with a polyclonal antibody  
against LBP

as well as flow cytometric analysis demonstrated its presence  
at the

parasite surface. After stimulation with phorbol-12-myristate-13-acet  
ate (PMA), U937 cells exhibited the ability to adhere to laminin

and LBP

specifically inhibited this adhesion. The reduced parasite adhesion  
after

tunicamycin treatment suggested the importance of sugar residues  
in cell

adhesion. Although co-administration of either laminin or LBP or  
anti LBP

antibody reduced parasite virulence, resulting in a lower  
level of

infection in the BALB/c mouse model, an in vitro  
macrophage

culture-enhanced level of infection was observed in the  
case of

laminin-coated parasites. The results collectively suggest a role  
for LBP

in the interaction of the parasite with extracellular matrix elements, which may constitute a basis for the homing of the parasite to its physiological address.

Record Date Created: 20010702

Record Date Completed: 20011205

10/7/52 (Item 1 from file: 155)  
DIALOG(R)File 155:MEDLINE(R)  
(c) format only 2009 Dialog. All rts. reserv.

08737821 PMID: 2450920

Monoclonal antibodies specific for the amastigote stage of *Leishmania pifanoi*. I. Characterization of antigens associated with stage- and species-specific determinants.

Pan A A; McMahon-Pratt D

Department of Epidemiology and Public Health, Yale University School of Medicine, New Haven, CT 06510.

Journal of immunology (Baltimore, Md. - 1950) (UNITED STATES)

Apr 1

1988, 140 (7) p2406-14, ISSN 0022-1767--Print Journal Code: 2985117R

Contract/Grant No.: AI-07136; AI; NIAID NIH HHS United States; AI-23004;

AI; NIAID NIH HHS United States

Publishing Model Print

Document type: Journal Article; Research Support, Non-U.S. Gov't;

Research Support, U.S. Gov't, P.H.S.

Languages: ENGLISH

Main Citation Owner: NLM

Record type: MEDLINE; Completed

Eight mAb were produced against membrane-enriched preparations of

*Leishmania pifanoi* amastigotes either grown in axenic culture (P-1 through P-6) or isolated from macrophage cell culture (P-7 and P-8). Two

mAb produced against promastigote membranes (P-9 and P-10) were found to be specific against this stage. Antibodies P-1 through P-8 on analysis

by radioimmune binding only reacted with determinants present on

amastigotes. mAb P-2, P-4, and P-8 also reacted with *Leishmania amazonensis* amastigotes but not promastigotes. No cross-reactions were found on any other species of *Leishmania* or with membranes of *Trypanosoma cruzi* epimastigotes or amastigotes. An indirect

immunofluorescence assay using mAb P-1 through P-8 confirmed the stage

specificity and binding to *L. pifanoi* axenically grown amastigotes, amastigotes within infected hamster tissue, and amastigotes within J774.1 macrophages. When Western blot analysis of amastigote membranes was conducted, one distinct group of molecules associated with *L. pifanoi*-specific determinants was identified. mAb P-1, P-3, P-5, P-7, and P-8 bound to molecules Mr 43 and 34 kDa. Promastigote-specific mAb P-9 recognized a diffuse pattern from 88 to greater than 200 kDa, and mAb P-10 localized a second class of proteins with Mr 53 kDa. On immunoprecipitation of solubilized [35S]methionine-labeled amastigotes, mAb P-2 recognized a doublet of Mr 35 and 33 kDa and another doublet at Mr 17.5 and 13.5 kDa. mAb P-4 and P-7 each precipitated a band at Mr 34 kDa. These studies indicate that antigenically the axenically cultured amastigote is closely related to macrophage-derived amastigote. These mAb and/or purified protein Ag may be useful in studying stage differentiation, monitoring transformation, and for further taxonomic, epidemiologic, and immunologic studies of New World leishmaniasis.

Record Date Created: 19880426

Record Date Completed: 19880426

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S2	51	RD S1 (unique items)
S3	175	AMAZONENSIS AND ((PROMASTIGOTE AND SURFACE AND ANTIGEN?) OR
		PSA OR ESA)
S4	55	RD S3 (unique items)
S5	177478	((PROMASTIGOTE AND SURFACE AND ANTIGEN?) OR PSA OR
		ESA)
S6	1112	S5 AND (LEISHMANIA OR AMAZONENSIS)
S7	335	RD S6 (unique items)
S8	298	S7 NOT PY>2004
S9	0	S8 AND 42.5
S10	52	S8 AND KDA

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Set	Items	Description
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S2	51	RD S1 (unique items)
S3	175	AMAZONENSIS AND ((PROMASTIGOTE AND SURFACE AND ANTIGEN?) OR

		PSA OR ESA)
S4	55	RD S3 (unique items)
S5	177478	((PROMASTIGOTE AND SURFACE AND ANTIGEN?) OR PSA OR
ESA)		
S6	1112	S5 AND (LEISHMANIA OR AMAZONENSIS)
S7	335	RD S6 (unique items)
S8	298	S7 NOT PY>2004
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